

WEST Search History

DATE: Friday, January 28, 2005

<u>Hide?</u>	<u>Set Name</u>	<u>Query</u>	<u>Hit Count</u>
	<i>DB=PGPB,USPT,USOC,EPAB,JPAB,DWPI; PLUR=YES; OP=OR</i>		
<input type="checkbox"/>	L15	L14 and mexicana	71
<input type="checkbox"/>	L14	L13	1130
<input type="checkbox"/>	L13	L12 and lysate	1130
<input type="checkbox"/>	L12	L11 and kit	1743
<input type="checkbox"/>	L11	L10 and leishmania	2625
<input type="checkbox"/>	L10	L9 and l8	646160
<input type="checkbox"/>	L9	skin test	1890731
<input type="checkbox"/>	L8	dextran free	3145409
<input type="checkbox"/>	L7	L6 and pressure	316
<input type="checkbox"/>	L6	L5 and diagnosis	400
<input type="checkbox"/>	L5	L4 and lysate	522
<input type="checkbox"/>	L4	L3 and phenol	659
<input type="checkbox"/>	L3	L2 and free	1313
<input type="checkbox"/>	L2	L1 and dextran	1396
<input type="checkbox"/>	L1	leishmania	3803

END OF SEARCH HISTORY

[First Hit](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)[Generate Collection](#)[Print](#)

L15: Entry 27 of 71

File: PGPB

Nov 14, 2002

PGPUB-DOCUMENT-NUMBER: 20020169285

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020169285 A1

TITLE: Leishmania antigens for use in the therapy and diagnosis of leishmaniasis

PUBLICATION-DATE: November 14, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Reed, Steven G.			US	
Campos-Neto, Antonio			US	
Webb, John R.			US	
Dillon, David C.			US	

US-CL-CURRENT: 530/350

CLAIMS:

1. An isolated polypeptide comprising an immunogenic portion of a Leishmania antigen or a variant thereof, wherein said antigen comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 2, 4, 20, 22, 24, 26, 36-38, 41, 50-53, 82, 104, 106, 108, 110, 112, 118-122, 134 and 135, and variants thereof.
2. An isolated antigenic epitope of a Leishmania antigen comprising an amino acid sequence selected from the group consisting of: SEQ ID NO: 43, 56, 57 or 58.
3. An isolated polypeptide comprising at least two contiguous antigenic epitopes according to claim 2.
4. An isolated polynucleotide comprising a DNA sequence encoding a polypeptide according to any one of claims 1 and 3.
5. The polynucleotide of claim 4, wherein the DNA sequence is selected from the group consisting of: SEQ ID NO: 1, 3, 19, 21, 23, 25, 29-31, 34, 45-48, 74, 102, 103, 105, 107, 109, 111, 113-117 and 129-133.
6. A recombinant expression vector comprising a polynucleotide according to claim 5.
7. A host cell transformed with an expression vector according to claim 6.
8. The host cell of claim 7 wherein the host cell is selected from the group consisting of E. coli, yeast and mammalian cells.
9. A fusion protein comprising at least two polypeptides according to any one of claims 1 and 3.
10. A fusion protein comprising at least two antigenic epitopes according to claim 2.

11. A pharmaceutical composition comprising a polypeptide according to any one of claims 1 and 3, and a physiologically acceptable carrier.
12. A pharmaceutical composition comprising a fusion protein according to any one of claims 9 and 10, and a physiologically acceptable carrier
13. An immunogenic composition comprising a polypeptide according to any one of claims 1 and 3 and an immunostimulant.
14. An immunogenic composition according to claim 13 further comprising a delivery vehicle.
15. The immunogenic composition of claim 14, wherein the delivery vehicle is a biodegradable microsphere.
16. An immunogenic composition comprising a polynucleotide according to claim 4 and an immunostimulant.
17. An immunogenic composition comprising a fusion protein according to any one of claims 9 and 10 and an immunostimulant
18. A method for inducing protective immunity against leishmaniasis in a patient comprising administering a pharmaceutical composition according to any one of claims 11 and 12.
19. A method for inducing protective immunity against leishmaniasis in a patient comprising administering an immunogenic composition according to any one of claims 13, 16 and 17.
20. A method for detecting Leishmania infection in a patient, comprising: (a) contacting dermal cells of the patient with a pharmaceutical composition according to any one of claims 11 and 12; and (b) detecting an immune response on the patient's skin.
21. The method of claim 20, wherein the immune response is induration.
22. A diagnostic kit comprising: (a) a pharmaceutical composition according to any one of claim 11 and 12; and (b) apparatus sufficient to contact dermal cells of a patient with the pharmaceutical composition.
23. The composition of claim 17, wherein the immunostimulant is selected from the group consisting of: aminoalkyl glucosaminide 4-phosphates; monophosphoryl lipid A; and 3-de-O-acylated monophosphoryl lipid A.

[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

Print

L15: Entry 27 of 71

File: PGPB

Nov 14, 2002

PGPUB-DOCUMENT-NUMBER: 20020169285

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020169285 A1

TITLE: Leishmania antigens for use in the therapy and diagnosis of leishmaniasis

PUBLICATION-DATE: November 14, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Reed, Steven G.			US	
Campos-Neto, Antonio			US	
Webb, John R.			US	
Dillon, David C.			US	

APPL-NO: 09/ 991496 [PALM]

DATE FILED: November 20, 2001

RELATED-US-APPL-DATA:

Application 09/991496 is a continuation-in-part-of US application 09/874923, filed June 4, 2001, PENDING

Application 09/874923 is a continuation-in-part-of US application 09/639206, filed August 14, 2000, PENDING

Application 09/639206 is a continuation-in-part-of US application 09/565501, filed May 5, 2000, PENDING

Application 09/565501 is a continuation-in-part-of US application 09/551974, filed April 14, 2000, PENDING

Application 09/551974 is a continuation-in-part-of US application 09/183861, filed October 30, 1998, US Patent No. 6365165

Application 09/183861 is a continuation-in-part-of US application 09/022765, filed February 12, 1998, US Patent No. 6375955

Application 09/022765 is a continuation-in-part-of US application 08/920609, filed August 27, 1997, PENDING

Application 08/920609 is a continuation-in-part-of US application 08/798841, filed February 12, 1997, PENDING

Application 08/798841 is a continuation-in-part-of US application 08/533669, filed September 22, 1995, US Patent No. 5834592

INT-CL: [07] C07 K 1/00, C07 K 14/00, C07 K 17/00

US-CL-PUBLISHED: 530/350

US-CL-CURRENT: 530/350

REPRESENTATIVE-FIGURES: NONE

ABSTRACT:

Compositions and methods for preventing, treating and detecting leishmaniasis and stimulating immune responses in patients are disclosed. The compounds provided include polypeptides that contain at least an immunogenic portion of one or more

Leishmania antigens, or a variant thereof. Vaccines and pharmaceutical compositions comprising such polypeptides, or polynucleotides encoding such polypeptides, are also provided and may be used, for example, for the prevention and therapy of leishmaniasis, as well as for the detection of Leishmania infection.

REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of U.S. patent application Ser. No. 09/874,923 filed Jun. 4, 2001, which is a continuation-in-part of U.S. patent application Ser. No. 09/639,206 filed Aug. 14, 2000, which is a continuation-in-part of U.S. patent application Ser. No. 09/565,501 filed May 5, 2000, which is a continuation-in-part of U.S. patent application Ser. No. 09/551,974 filed Apr. 14, 2000, which is a continuation-in-part of U.S. patent application Ser. No. 09/183,861, filed Oct. 30, 1998 (allowed), which is a continuation in part of U.S. patent application Ser. No. 09/022,765, filed Feb. 12, 1998 (allowed), which is a continuation-in-part of U.S. patent application Ser. No. 08/920,609, filed Aug. 27, 1997, which is a continuation-in-pan of U.S. patent application Ser. No. 08/798,841, filed Feb. 12, 1997, which is a continuation-in-part of U.S. patent application Ser. No. 08/533,669, filed Sep. 22, 1995, now U.S. Pat. No. 5,834,592, and are incorporated in their entirety herein by reference.

[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

Print

L15: Entry 63 of 71

File: USPT

Nov 10, 1998

US-PAT-NO: 5834592

DOCUMENT-IDENTIFIER: US 5834592 A

TITLE: Leishmania antigens for use in the therapy and diagnosis of Leishmaniasis

DATE-ISSUED: November 10, 1998

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Reed; Steven G.	King	WA		
Campos-Neto; Antonio	King	WA		
Webb; John R.	King	WA		
Dillon; Davin C.	King	WA		
Skeiky; Yasir A. W.	King	WA		

US-CL-CURRENT: 530/350; 424/184.1, 424/269.1, 530/364, 530/806, 930/210

CLAIMS:

We claim:

1. An isolated polypeptide comprising an immunogenic portion of a Leishmania antigen having the amino acid sequence recited in SEQ ID NO: 4, or a variant of said antigen that differs only in conservative substitutions, modifications or combinations thereof.
2. The polypeptide of claim 1, comprising amino acids 1-175 of SEQ ID NO:4.

[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#) [Previous Doc](#) [Next Doc](#) [Go to Doc#](#)

Generate Collection

Print

L15: Entry 22 of 71

File: PGPB

Apr 17, 2003

PGPUB-DOCUMENT-NUMBER: 20030072714

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20030072714 A1

TITLE: Microfluidized leishmania lysate and methods of making and using thereof

PUBLICATION-DATE: April 17, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Magill, Alan J.	Kensington	MD	US	
Stiteler, John M.	Springfield	VA	US	
Grogl, Max	Columbia	MD	US	
Rowton, Edgar D.	College Park	MD	US	
Eckels, Kenneth H.	College Park	MD	US	
Ballou, William R.	Silver Spring	MD	US	

APPL-NO: 09/ 975020 [PALM]

DATE FILED: October 12, 2001

INT-CL: [07] A61 K 49/00, G01 N 33/53, G01 N 33/569

US-CL-PUBLISHED: 424/9.81; 435/7.22

US-CL-CURRENT: 424/9.81; 435/7.22

REPRESENTATIVE-FIGURES: NONE

ABSTRACT:

Disclosed herein are microfluidized lysate preparations of Leishmania parasites and methods of making thereof. Also disclosed are methods of using the microfluidized lysate preparations in skin test antigen assays as well as kits comprising the microfluidized lysate preparations. The microfluidized lysate preparations are made under current good manufacturing practice and may therefore be standardized and such preparations may be produced with consistently.

[Previous Doc](#) [Next Doc](#) [Go to Doc#](#)

[First Hit](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

Print

L15: Entry 12 of 71

File: PGPB

Jan 8, 2004

PGPUB-DOCUMENT-NUMBER: 20040005326

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040005326 A1

TITLE: Leishmania vaccine

PUBLICATION-DATE: January 8, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Mottram, Jeremy Charles	Bearsden		GB	
Coombs, Graham Herbert	Glasgow		GB	

US-CL-CURRENT: 424/184.1

CLAIMS:

1. Use of a mutant Leishmania in the preparation of a vaccine, wherein the mutant Leishmania comprises at least one defective cysteine proteinase gene type, such that the mutant Leishmania is substantially incapable of expressing a functionally active form of said at least one cysteine proteinase.
2. Use of a mutant Leishmania according to claim 1 wherein the mutant Leishmania comprises two or more defective cysteine proteinases.
3. A vaccine formulation comprising a mutant Leishmania, said mutant Leishmania comprising at least one defective cysteine proteinase gene type, such that the mutant Leishmania is substantially incapable of expressing a functionally active form of said at least one cysteine proteinase.
4. A vaccine formulation according to claim 3 wherein the mutant Leishmania comprises two or more defective cysteine proteinases.
5. A vaccine formulation according to either of claims 3 or 4 wherein the mutant Leishmania is selected from any species of Leishmania including L. braziliensis, L. peruviana, L. guyanensis, L. mexicana, L. major, L. amazonensis, L. infantum, L. chagasi and L. donovani.
6. A vaccine formulation according to claim 5 wherein the mutant Leishmania is a L. mexicana mutant and said defective cysteine proteinase gene(s) is/are selected from cpa, cpb and/or cpc.
7. A vaccine formulation according to claim 6 wherein the L. mexicana mutant is a cpb single null mutant.
8. A vaccine formulation according to claim 6 wherein the L. mexicana mutant is a cda/cpb double null mutant.
9. A vaccine formulation according to claim 5 wherein the mutant Leishmania is a L. infantum mutant and said defective proteinase gene(s) is/are selected from cpa and/or cpb.

h e b b g e e e f c e

e ge

10. A vaccine formulation according to claim 9 wherein the L. infantum cpa is identifiable from the sequence shown in FIG. 8 and/or the cpb is identifiable from the sequence as shown in FIG. 10.

11. A vaccine formulation according to any preceding claim wherein the cysteine proteinase gene has been modified by a deletion, insertion, substitution or rearrangement such that said cysteine proteinase gene(s) is/are substantially incapable of expressing a functionally competent cysteine proteinase.

12. A vaccine formulation according to claim 11 wherein said cysteine proteinase gene has been modified by deletion of all or a portion of said cysteine proteinase gene.

13. A vaccine formulation according to claim 12 wherein a gene or gene fragment capable of expressing a polypeptide selected from polypeptides which augment an immune response and marker polypeptides is inserted with a gap generated by deletion of all or the portion of said cysteine proteinase gene.

14. A vaccine formulation according to claim 13 wherein the polypeptide is a cytokine.

15. A vaccine formulation according to claim 13 wherein at least one copy of said cysteine proteinase gene has been modified such that a substantially inactive form of a cysteine proteinase polypeptide is expressed.

16. A vaccine formulation according to any preceding claim wherein the mutant Leishmania is a drug resistant marker-free mutant.

17. A vaccine formulation according to any preceding claim for eliciting at least a cellular immune response.

18. A vaccine formulation according to claim 17 wherein the cellular immune response is a Th1 cell response.

19. A vaccine formulation according to any preceding claim further comprising an adjuvant and/or cytokine.

20. A vaccine formulation according to any preceding claim further comprising at least one disfunctional cysteine proteinase, wherein said disfunctional cysteine proteinase is substantially enzymatically inactive, but which is antigenic or immunogenic.

21. A method of vaccinating against Leishmania which comprises administering to an animal an effective, non-toxic amount of a vaccine formulation according to any one of claims 3-20.

22. A method according to claim. 21 wherein the method comprises parenteral administration.

23. Use of a mutant Leishmania in the manufacture of a vaccine for the prophylaxis and/or treatment of Leishmaniasis, wherein the mutant comprises at least one defective cysteine proteinase gene type, such that the mutant Leishmania is substantially incapable of expressing a functionally active form of said at least one cysteine proteinase.

24. A pharmaceutical formulation comprising a vaccine according to any one of claims 3-20 together with a carrier or excipient.

[Previous Doc](#)

[Next Doc](#)

[Go to Doc#](#)

[First Hit](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)[Generate Collection](#)[Print](#)

L15: Entry 30 of 71

File: PGPB

Mar 7, 2002

DOCUMENT-IDENTIFIER: US 20020028215 A1

TITLE: NOVEL VACCINES AND PHARMACEUTICAL COMPOSITIONS USING MEMBRANE VESICLES OF MICROORGANISMS, AND METHODS FOR PREPARING SAME

Summary of Invention Paragraph:

[0004] Conventional prophylactic treatments for infectious diseases are also becoming increasingly ineffective with the emergence of resistant mutant strains of infectious agents. Vaccines for the prophylaxis of infectious diseases have been developed which incorporate whole attenuated organisms, cell lysates, culture supernatants or extracts of the infectious agents. There has been considerable interest in improving existing vaccines since they typically contain fractions having physical or chemical characteristics which result in toxicity or undesired immune responses.

Summary of Invention Paragraph:

[0015] The invention also provides a method for screening for an immunogenic antigen of a pathogen comprising (a) providing a membrane vesicle having a test antigen associated with its surface; (b) vaccinating an animal with the membrane vesicle; and (c) challenging the animal with the pathogen to determine if the test antigen provides protection against the pathogen.

Detail Description Paragraph:

[0069] The microorganisms which produce membrane vesicles described herein may also be transfected with one or more nucleotide sequences encoding exogenous proteins in order to provide membrane vesicles have exogenous proteins incorporated into the membrane vesicles or associated with their surface. For example, the exogenous proteins include antigens which are associated with infectious diseases caused by infectious agents which do not produce membrane vesicles including viruses such as human immunodeficiency virus (HIV), influenza (neuraminidase/haemagglutinin), adenovirus, Herpes simplex, measles, simian immunodeficiency virus; fungi such as Histoplasma capsulatum, Cryptococcus neoformans, Blastomyces dermatidis, Candida albicans; protozoa such as Leishmania mexicana, Plasmodium falciparum and Toxoplasma gondii; and, gram-positive bacteria such as Streptococcus mutans, and S. pneumoniae (cell wall antigens). Microorganisms transfected with such antigens may be used to produce membrane vesicles which may be used as vaccines against the infectious agent. The microorganism may also be transfected with a nucleotide sequence encoding an exogenous protein having a known therapeutic or regulatory activity such as hormones preferably insulin, blood clotting factor VIII, growth hormones, hirudin, cytokines such as gamma interferon, tumor necrosis factor, IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, GM-CSF, CSF-1, and G-CSF. Membrane vesicles containing therapeutic or regulatory proteins may be used to deliver the proteins to a host. The microorganisms may also be transfected with proteins which facilitate targeting of a membrane vesicle having the proteins associated with their surfaces to specific target tissues or cells. For example, tumor-associated antigens, CD.sub.4 proteins on T-helper cells, and gp120 in HIV.

Detail Description Paragraph:

[0090] In accordance with another embodiment of the invention, a vaccine against infectious diseases caused by an infectious agent which does not produce membrane vesicles is provided which comprises a carrier strain having a membrane vesicle from a microorganism integrated into the cell surface of the carrier strain, wherein the membrane vesicle has an amount of an antigen associated with its surface which is effective to provide protection against the infectious agent. The vaccines may be used for the prophylaxis or active immunization and treatment of

infectious diseases caused by microorganisms including viruses such as human immunodeficiency virus (HIV), influenza (neuraminidase/haemagglutinin), adenovirus, Herpes simplex, measles, simian immunodeficiency virus; fungi such as Histoplasma capsulatum, Cryptococcus neoformans, Blastomyces dermatidis, Candida albicans; protozoa such as Leishmania mexicana, Plasmodium falciparum and Toxoplasma gondii; and, gram-positive bacteria such as Streptococcus mutans, and S. pneumoniae. Therefore, the vaccines of the present invention may incorporate membrane vesicles with immunogenic antigens of these microorganisms.

Detail Description Paragraph:

[0114] Administration of an amount effective to have a bactericidal effect is defined as an amount effective, at dosages and for periods of time necessary to achieve the desired result. For example, an amount effective to have a bactericidal effect may vary according to factors such as the disease state, age, sex, and weight of the individual. Dosage regimes may be adjusted to provide the optimum therapeutic response. Amounts of membrane vesicles effective to have a bactericidal effect on a selected gram-negative and/ or gram-positive bacterial pathogen may be determined using conventional in vivo and in vitro tests (see zymogram systems as outlined in Bernadsky, G. et al. supra).

Detail Description Paragraph:

[0159] The invention also relates to a method of inserting nucleic acid molecules into a target cell which comprises encapsulating the nucleic acid in a membrane vesicle of a microorganism, and bringing the membrane vesicle in contact with the target cell whereby the nucleic acid molecule is inserted into the cell. Nucleic acid molecules which may be encapsulated in a membrane vesicle may be from eucaryotic or prokaryotic cells and they may be endogenous or exogenous to a microorganism that produces membrane vesicles. Examples of nucleic acid molecules which may be encapsulated in a membrane vesicle are nucleic acid molecules encoding (a) mammalian proteins such as hormones preferably insulin, blood clotting factor VIII, growth hormones, hirudin, cytokines, and a normal copy of the cystic fibrosis transmembrane conductance regulator (CFTR); (b) viral antigens such as HIV glycoprotein, hepatitis B surface antigens, influenza antigens; fungal antigens for example from Histoplasma capsulatum, Cryptococcus neoformans, Blastomyces dermatidis, Candida albicans; and (c) protozoal antigens for example from Leishmania mexicana, Plasmodium falciparum and Toxoplasma gondii.

Detail Description Paragraph:

[0169] Antibiotic Susceptibility Test.

Detail Description Paragraph:

[0176] Proteinases were resolved by SDS-PAGE using the method of Matsumoto et. al. (Invest. Ophthalmol. Vis. Sci. 34:1945-1953, 1993) with slight modifications. The separating gels used were 8% SDS gels containing a-casin or gelatin (Type A from bovine skin; Sigma) at a final concentration of 0.15%. The stacking gels consisted of 4% SDS gels without gelatin or casein. A 25 .mu.g protein sample from each preparation (without reducing agents) was loaded onto gels and was run at 4.degree. C. for 90 min. at 120 V. After electrophoresis, the gels were shaken at room temperature in a solution of 2.5% Triton-X100 for 45 min. to remove the SDS. Subsequently, the gels were incubated at 37.degree. C. in incubation buffer (50 mM Tris-HCl, pH 8.0, 5 mM CaCl.sub.2) with 5 mM EDTA for 18 h. The positions of the proteinases were identified after the gels were stained (0.5% Coomassie brilliant blue R-250, 10% acetic acid, 40% methanol) for 2 h and clear bands were identified.

Detail Description Paragraph:

[0180] The DNA content in MVs was quantitated using an assay developed by the Pierce Chem. Co. (Pierce, Rockford, Ill.) according to the manufacturer's instructions. Briefly, 20 .mu.g of protein from MVs in 50 .mu.l assay buffer (0.1 M NaCl, 10 mM EDTA, 10 mM Tris, pH 7.0) was lysed with 50 .mu.l of extraction solution (0.1 M NH.sub.4OH, 0.2% Triton X-100). A standard curve for DNA was prepared with calf thymus DNA (0-150 ng/ml) (provided with the assay kit) in 200 mM NaCl, 20 mM EDTA, pH 7.0, 0.05 NH.sub.4OH, 0.01% Triton X-100). To each sample, 1.5 ml of fluorescent dye (200 .mu.g/ml) (Bisbenzimidazole) was added, the tubes

were capped quickly, mixed and fluorescence was measured in a Hitachi F-2000 fluorescence spectrophotometer with excitation and emission wave lengths set at 350 and 455 nm, respectively (10-nm slit width), and yielded values for total DNA/mg of protein. Experiments were also performed on MVs without their being treated with extraction solution. For some experiments, the intact MVs and purified DNA were treated with pancreatic DNase 1 (1.0g/ml; Sigma).

Detail Description Paragraph:

[0181] Preparation of Cell Lysates, Supernatants and MVs for Enzyme Assays.

Detail Description Paragraph:

[0182] Membrane-filtered supernatants, before and after harvesting MVs, were concentrated 10-fold in a Concentrator evaporator (jouan, Winchester, Va.). Washed whole cells or MVs, were sonicated for 2 min. with 0.1 % v/v toluene to release intracellular enzymes in a sonic bath (Bransonic Ultrasonic Corporation, Iainburg, Conn.). Protein concentrations of samples were determined with the micro BCA reagent kit (Pierce). Whole cells and MVs (both at a 20 .mu.g protein concentration) or concentrated supernatants (50 .mu.l) were assayed for enzyme activity.

Detail Description Paragraph:

[0189] FIG. 1 shows thin sections of H 103 cells showing the formation of vesicles (solid arrowheads) and free MVs in growth medium (open arrow); (A), Control and (B), exposed to 4.times. MIC of gentamicin. A larger number of Mvs are formed from the cell surface of bacteria exposed to gentamicin than from unreacted cells. Electron dense material has been trapped in the developing and free vesicles. Bar=100 nm.

Detail Description Paragraph:

[0190] FIG. 1 shows thin-sections of P. aeruginosa either treated with gentamicin or untreated. The untreated control cells (FIG. 1(A)) represent "natural" cultured cells and possessed intact cell envelopes, with several membrane blebs emanating from each cell surface or free in the environment. Cells that were exposed to gentamicin formed many more blebs (FIG. 1(B)) than those seen in untreated cells. At a gentamicin concentration of 8 .mu.g/ml, this increase in blebbing was visible after approximately 1 min of antibiotic incubation. Examination of intact isolated purified blebs from both natural and gentamicin-treated cultures in negative stains showed that, although they were partially collapsed, many were filled with a particulate substance (FIG. 2(A) is an electron micrograph showing a negative stain of intact gentamicin-MVs (g-MVs)). This was better shown and confirmed with thin sections (FIG. 2(B) shows an electron micrograph of a thin section of intact g-MVs). The diameter of the vesicles from both untreated and gentamicin-treated cells varied between 50 nm to 150 nm when measured in thin sections; however, when measurements of g-MVs were averaged, the g-MVs were found to be slightly larger than natural MVs (n-MVs), with a mean diameter of 100 nm as opposed to 80 nm for n-MVs. Thin sections proved the vesicles to have a bilayer structure (FIG. 2(B)). No external material was seen by any TEM technique, thereby suggesting the isolated vesicles were free from particulate cellular debris.

Detail Description Paragraph:

[0194] The protein profiles of whole cell lysates, OMPs extracted from whole cells and MVs from untreated or treated cells were compared by SDS-PAGE. FIG. 3 shows SDS-PAGE profiles of n-MV, g-MV, outer membrane proteins (OMP), and control whole cells (WCC) in a 12% polyacrylamide gel stained with Comassie brilliant blue. Each lane contains 25 .mu.g of total protein from the indicated samples. Molecular masses (in kilodaltons) are indicated on the left.

Detail Description Paragraph:

[0195] The n-MVs and g-MVs contained much fewer protein bands than the OMPs extracted from whole cells or whole cell lysates. The banding patterns of n-MVs and g-MVs were very similar, but not identical; both types of Mvs appeared to have lost several bands which were normally present in whole cell lysates and the OMP samples. Some of the prominently stained bands from both vesicle preparations included .about.70 kD, 40 kD and 20 kD proteins. Trace amounts of an -35 kD protein

was detected in g-MVs but not in n-MVs.

Detail Description Paragraph:

[0200] Tables 2 and 3 illustrate the enzymatic activities in cellular extracts, MVs and culture supernatants, from cultures which were treated with gentamicin or untreated. Both types of vesicles exhibit PLC activity, as measured spectrophotometrically by the hydrolysis of p-nitrophenylphosphorylcholine, indicating that the enzyme is associated with the MVs. To evaluate the PLC activity in the supernatants, the enzyme activity was assayed before and after the removal of vesicles from cell-free culture supernatants. Removal of vesicles from gentamicin-treated cultures resulted in an 83% reduction in enzyme activity as compared to a 68% decrease in untreated cultures (Table 3). This suggests that the majority of PLC secreted into the external environment is indeed concentrated in the vesicles. The observed difference in enzyme activity between the two cultures is due to the fact that the amount of vesicles per unit mass is greater in gentamicin-exposed cultures than in untreated cultures, hence a higher percentage of PLC activity is removed with the vesicles. It has been reported previously that *P.aeruginosa* produces and excretes two distinct PLCs with similar activities; each is capable of acting on the substrate, phosphatidylcholine (Shortridge, V. D. et al., Mol Microbiol. 6:863-871, 1992). Although both PLCs hydrolyse this substrate, one is hemolytic (PLC-H) for sheep and human erythrocytes and is heat labile, while the other (PLC-N) is not. Additionally, PLC-H can hydrolyse sphingomyelin, but not phosphatidylserine, whereas PLC-N hydrolyses phosphatidylserine but not sphingomyelin (Bergmann U., et al., Infect. Immun. 57:2187-2195, 1989; Berk, R. S. Infect. Immun. 55:1728-1730, 1987; and, Vasil, M. L. et al., Antibiot. Chemother. 44:34-47, Karger, Basel, 1991). The MVs were examined for hemolytic activity on sheep blood agar plates as well as spectrophotometrically on sheep blood cells, and it was found that both types of MVs were positive. No attempt was made to differentiate between PLC-H and PLC-N in the study.

Detail Description Paragraph:

[0202] *P. aeruginosa* secretes several proteases (Hastie, A., et al., Infect. Immun. 40:506-513, 1983; Kessler, E., et al., J. Biol. Chem. 268:7503-7508; Lazduski, A. J., et al., Biochimie 72:147-156, 1990; and, Wretling, B., and O. R. Pavlovskis, Rev. Infect. Dis. 5:S998-1004, 1983). The secretion of elastase and protease was examined in strain ATCC 19660 since the amounts of both enzymes produced by this strain was found to be larger than those for strain H 103. No appreciable amount of proteolytic or elastolytic activity was detected in toluene cellular extracts, indicating the lack of intracellular accumulation of active enzymes (Table 2). This observation was in agreement with earlier work (Duong, F. et al, Gene 121:47-54, 1992; Guzzo, J., et al, J.Bacteriol. 173:5290-5297, 1991; Hamood, A.N. et al., Infect. Immun. 60:510-517, 1992; Hastie, A. et al., Infect Immun. 40:506-513, 1983; Kessler, E., et al., J. Biol. Chem. 268:7503-7508, 1993; and Lazduski, A. J., et al., Biochimie 72:147-156, 1990). Examination of both types of MVs for protease activity demonstrated the association of active enzyme. On removal of vesicles from cell-free culture supernatants, the total protease activity dropped by 18% in untreated culture supernatants and 25% in gentamicin-treated culture supernatants. Since an appreciable amount of activity could also be detected in culture supernatants following the removal of vesicles from cell-free medium, the enzyme is probably released from cells in both soluble and vesicle-associated forms. In contrast, elastolytic activity was detected exclusively in culture supernatants and was not affected by removal of vesicles from cell-free culture supernatants. Previous studies have demonstrated that the enzyme is secreted as a proenzyme that becomes active only as it is released into the supernatant (Guzzo, J. et al., J. Bacteriol. 173:5290-5297, 1991; and Kessler, E. et al., J Biol. Chem. 268:7503-7508, 1993). For this reason, even if the proenzyme is present in MVs, it would not be detectable by its enzyme activity.

Detail Description Paragraph:

[0208] As judged by the location of gold particles on thin sections, a uniform distribution of the enzyme in the cytoplasm is clearly seen. Interestingly, it can also be seen that cytoplasm is streaming into a forming vesicle. Budding and free vesicles were labelled to the same extent with gold particles, demonstrating that PLC is entrapped within both types of MVs. This was in good agreement with the

h e b b g e e f c e e ge

biochemical demonstration of the enzyme activity in vesicle preparations (Tables 2 and 3). Immunogold labelling for the localization of alkaline phosphatase in thin-sections of intact cells and Mvs demonstrated that the majority of the enzyme was located in the envelope, particularly in the periplasm and outer membrane (FIG. 7). MVs were labelled on the membrane and on the luminal material attached to the membrane. g-MVs and n-MVs were labelled to approximately the same extent, confirming the result of the enzymatic assay (Table 2).

Detail Description Paragraph:

[0212] In particular, FIG. 9 shows Western immunoblot analysis of samples with antibodies to (A) elastase and (B) alkaline protease. Whole cell extracts from untreated control cells (WCC) or gentamicin-treated cells (WCG), n-MVs and g-MVs and cell-free supernatants after removal of MVs from untreated (n-sup) or gentamicin-treated (g-sup) cultures are shown. Lane P, contained purified elastase. Each lane contains 25 .mu.g of protein or 10 .mu.l of concentrated n-sup or g-sup. FIG. 9(C) shows proteinase present in MVs tested by gelatin zymography. Both n-MVs and g-MVs demonstrated three major bands (M.sub.r.about.33, 35 and 135 kDa) having proteolytic activity. Molecular masses (in kilodaltons) are indicated of the left and right.

Detail Description Paragraph:

[0218] Because the Mvs were isolated from early stationary phase growth cultures, it was also possible that some of the DNA was derived from lysed cells (within each culture) which had bound to the outer face of the vesicles. This could especially be true of g-MVs. Control experiments conducted with exhaustive treatment of MVs with pancreatic DNase showed that this was not the case. DNase-treated n- and g-MVs possessed amounts of DNA similar to those in Table 4. These control experiments also confirmed that the MVs were intact, since the DNA of the MVs was protected from the external enzyme. When in similar experiments containing Mvs and free DNA were treated with pancreatic DNase, and ethidium-bromide gel electrophoresis was performed, the external DNA was shown to be digested whereas the MV DNA remained intact.

Detail Description Paragraph:

[0308] In the *P. aeruginosa* system, although most extracellular autolytic activity is associated with MVs, some soluble activity can also be demonstrated. Therefore, once MVs are removed from the spent culture medium, there is still some residual peptidoglycan degrading activity. If *E. coli* is incubated with this spent liquor or the MVs lysate, there is no drop in viability indicated that the bacterium's outer membrane is an impermeable barrier to the soluble autolysins, this emphasizes the importance of the MV's bilayered membrane in directly entrapped autolysins to the *E. coli* (or other gram-negative) peptidoglycan layer. Once the MVs adhere to the outer membrane, the MV's membrane and outer membrane must fuse together, emptying the MV luminal contents into the host periplasm where the *P. aeruginosa* autolysins hydrolyse the peptidoglycan layer of intact cell.

Detail Description Paragraph:

[0312] The effect of n-MVs, g-MVs and gentamicin (2.5.times.MIC of antibiotic for gentamicin sensitive strains or 100 .mu.g/ml of gentamicin for the 8803 strain) on the viability of D.sub.2C, DH5.alpha., PA01, and 8803 is shown in FIG. 23. *P. aeruginosa* PA01 is the parent strain from which the MVs are derived. It was exquisitely sensitive to both g-MVs and the free gentamicin at 2.5 MIC. There was a small but discernable loss of viability of *S. aureus*, *E. coli* and *P. aeruginosa* PA01 (FIGS. 23(a), (b) and (c) exposed to n-MVs. Even though peptidoglycan hydrolysis must have occurred (FIGS. 19 and 20), the loss in viability was less than expected. DH5.alpha. and PA01 remained at a constant cell number for the first 1-2 h, and this gradually increased by 5 h. The n-MV cell numbers were only slightly reduced to those of the control cultures. The small effect of n-MVs on these cultures was presumably due to a rapid replacement of lysed cells with newly divided cells or to a rapid replacement of hydrolyzed peptidoglycan with newly synthesized polymer so that lysis was inhibited.

Detail Description Paragraph:

[0335] Mouse Immunization: Six-to seven week old female BALB/c mice (in groups of

six) were immunized orally via a gavage tube, with 0.3 ml of one of the following test vaccines: (i) Ty21a (2 x 10^{sup}.8 CFU/ml); (ii) PA01 MVs (100 .mu.g protein/ml); (iii) M90T MVs (100 fig protein/ml); (iv) Ty21a (2.times.10.sup.8 CFU/ml)+M90T MVs (at 100 .mu.g protein/ml) (v) Ty21a (2.times.10.sup.8 CFU/ml)+PA01 MVs (100 .mu.g/ml); (vi) Ty21a (2.times.10.sup.8 CFU/ml)+PA01 MVs+M90T MVs (at 100 .mu.g protein/ml); and, (vii) a control group with 0.3 ml sterile PBS. All vaccines were suspended immediately before immunization in 3% NaHCO₃ in PBS at pH 8.0, and given four times at one week intervals. One week after the final immunization, mice were sacrificed, bled and the serum was collected. Bronchoalveolar washings were obtained as described in Guzman, C. A. et al, 1991, Infect. Immun. 59:4391-4397. Briefly, trachea and lungs were aspirated with 2.0 ml of ice-cold PBS containing 2 mM phenylmethylsulfonylfluoride (PMSF as a protease inhibitor) (Sigma Chemical Co., St. Louis, Mo.), three times to get an even distribution of the solution between each lung, before collecting the final fluid having a volume of 0.6-0.8 ml. Gut washes were obtained by washing the complete gut segment which was distal from the stomach with 1.5 ml ice-cold PBS-PMSF. The washings were centrifuged at 3000 x g for 10 min. at 4.degree. C. to remove cellular debris and stored at -20.degree. C. until tested.

Detail Description Paragraph:

[0341] By electron microscopy of thin sections isolated, purified MVs from *S. flexneri* strain M90T and *P. aeruginosa* strain PA01 were bilayered spherical vesicles ca. 80 nm in diameter and (most) were filled with a particulate substance. The protein profiles of whole cell lysates, OMPs (extracted for whole cells), and MVs from M90T and PA01 were compared by SDS-PAGE (FIG. 24(a)). The banding patterns of M90T MVs were similar, but not identical, to the corresponding OMPs from whole cells; the major .about.35 and 37 kDa. The prominently stained MV bands from PA01 included .about.-45 and 55-kDa proteins which were also prominent in the whole cell OMP fraction.

Detail Description Paragraph:

[0347] Serum or mucosal samples in which specific immunoglobulins could be detected by ELISA were next analysed by Western blotting to determine whether the induced antibodies were directed against LPS or protein antigens (FIG. 27). Samples were analysed for serum IgA, IgG and IgM and mucosal IgA with sample buffer or proteinase-K treated M90T or PA01 whole cells as the antigen. Proteinase-K treatment was used to de-proteinize the antigenic samples so that those antibodies directed against LPS in the body fluid could more easily be detected. Strong anti-LPS antibodies were evident for both M90T and PA01-specific LPS. The IgA, IgG, and IgM responses to the various vaccine constructs showed a similar trend to that previously seen in ELISA tests. The antibody response to M90T MVs was weak with barely detectable bands on Western blots (FIG. 26). Immunoblotting of non-deproteinized samples with serum, lung, or gut washes revealed several immunoreactive protein-specific antibody responses (arrows) to the PA01 (FIG. 27(b)) and M90T (FIG. 27(d)) vaccine constructs. The reactivity of immunoglobulin classes to the various vaccine constructs was similar to the ELISA results. On these immunoblots, the LPS-specific antibody response was also visible for both PA01 and M90T and, in fact, dominated the protein responses when the two overlapped. The spacing and banding patterns were more extensive and complicated in these immunoblots than those treated with proteinase-K implying that the immune response is to both LPS and protein antigens. The antibody response to the prominent 35-37kDa and 45-55 kDa protein bands of M90T and PA01 (FIG. 24(a)) became evident when LPS-specific antibodies were removed from body fluids by adsorbing them out with either M90T or PA01 LPS. The antibody response to the carrier strain, Ty21a, was mainly protein-specific (FIG. 27(d)). This was to be expected, since the growth conditions employed did not promote complex side chain LPS expression in Ty21a.

Detail Description Table CWU:

3TABLE 3 Enzyme activities in cell-free culture supernatants following the removal of MVs. % Activity remaining in cell-free supernatant after removal of MVs Enzyme
n-MV g-MV PLC 32 .+-. 8 17 .+-. 5 Alk. phosphatase 49 .+-. 7 52 .+-. 6 Elastase
98 .+-. 0.8 98 0.9 Protease 82 .+-. 1.9 75 .+-. 4 abbreviations as in Table 2.

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

Print

L15: Entry 36 of 71

File: USPT

Oct 28, 2003

US-PAT-NO: 6638517

DOCUMENT-IDENTIFIER: US 6638517 B2

TITLE: Leishmania antigens for use in the therapy and diagnosis of leishmaniasis

DATE-ISSUED: October 28, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Reed; Steven G.	Bellevue	WA		
Campos-Neto; Antonio	Bainbridge Island	WA		
Webb; John R.	Manotick	CA		
Dillon; Davin C.	Issaquah	WA		
Skeiky; Yasir A. W.	Bellevue	WA		
Bhatia; Ajay	Seattle	WA		
Coler; Rhea N.	Seattle	WA		
Probst; Peter	Seattle	WA		
Brannon; Mark	Seattle	WA		

US-CL-CURRENT: 424/269.1; 424/184.1, 424/191.1, 424/192.1, 424/265.1, 424/450,
424/85.2, 435/69.7, 514/12, 514/2, 514/44 , 530/300, 530/350, 536/23.1, 536/23.4

CLAIMS:

What is claimed is:

1. A method for inducing protective immunity against leishmaniasis in a patient comprising administering an immunogenic composition comprising a fusion protein and an immunostimulant, wherein the fusion protein comprises the amino acid sequence of SEQ ID NO:24.
2. The method of claim 1, wherein the fusion protein comprises the amino acid sequence of SEQ ID NO:95.
3. The method of any one of claims 1 and 2, wherein the immunostimulant is selected from the group consisting of: aminoalkyl glucosaminide 4-phosphates; monophosphoryl lipid A; and 3-de-O-acylated monophosphoryl lipid A.

[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

Print

L15: Entry 38 of 71

File: USPT

Aug 19, 2003

US-PAT-NO: 6607731

DOCUMENT-IDENTIFIER: US 6607731 B1

TITLE: Leishmania antigens for use in the therapy and diagnosis of leishmaniasis

DATE-ISSUED: August 19, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Reed; Steven G.	Bellevue	WA		
Campos-Neto; Antonio	Bainbridge Island	WA		
Webb; John R.	Manotick			CA
Dillon; Davin C.	Issaquah	WA		
Skeiky; Yasir A.	Bellevue	WA		
Bhatia; Ajay	Seattle	WA		
Coler; Rhea	Seattle	WA		
Probst; Peter	Seattle	WA		

US-CL-CURRENT: 424/269.1, 424/184.1, 424/185.1, 424/191.1, 424/192.1, 424/265.1,
514/12, 514/2, 514/46, 530/300, 530/350 , 536/23.1, 536/23.4

CLAIMS:

What is claimed is:

1. A fusion protein comprising the amino acid sequence of SEQ ID NO:24.
2. A fusion protein comprising an immunogenic portion of SEQ ID NO:24, wherein said immunogenic portion selectively binds to anti-Leishmania antibodies specific for SEQ ID NO: 24.

[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

First Hit Fwd RefsPrevious DocNext DocGo to Doc#

Generate Collection

Print

L15: Entry 51 of 71

File: USPT

Apr 23, 2002

US-PAT-NO: 6375955--
DOCUMENT-IDENTIFIER: US 6375955 B1 --TITLE: Leishmania antigens for use in the therapy and diagnosis of leishmaniasis

DATE-ISSUED: April 23, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Reed; Steven G.	Bellevue	WA		
Campos-Neto; Antonio	Bainbridge Island	WA		
Webb; John R.	Manotick			CA
Dillon; Davin C.	Issaquah	WA		
Skeiky; Yasir A. W.	Bellevue	WA		
Bhatia; Ajay	Seattle	WA		
Probst; Peter	Seattle	WA		

US-CL-CURRENT: 424/269.1; 424/184.1, 424/265.1, 530/300, 530/350, 930/210

CLAIMS:

What is claimed is:

1. An isolated polypeptide comprising an immunogenic portion of a Leishmania antigen, wherein said antigen comprises an amino acid sequence of SEQ ID NO: 82.
2. A pharmaceutical composition comprising a polypeptide according to claim 1, and a physiologically acceptable carrier, wherein the polypeptide is present in an amount effective for the treatment of Leishmaniasis.
3. A pharmaceutical composition according to claim 2, further comprising soluble Leishmania antigens.
4. A pharmaceutical composition according to claim 2, further comprising a K39 Leishmania antigen.
5. A pharmaceutical composition comprising a polypeptide and a physiologically acceptable carrier, the polypeptide comprising an immunogenic portion of a Leishmania antigen, wherein said antigen comprises an amino acid sequence selected from the group consisting of SEQ ID NO:80, 81, and 83-87, the polypeptide being present in an amount effective for the treatment of Leishmaniasis.
6. A pharmaceutical composition according to claim 5 further comprising a K39 Leishmania antigen.
7. An isolated polypeptide comprising an amino acid sequence of SEQ ID NO:61, wherein said polypeptide comprises an immunogenic portion of a Leishmania antigen, said immunogenic portion of Leishmania antigen selectively binds to

anti-leishmania antibodies.

8. An isolated polypeptide comprising an amino acid sequence of SEQ ID NO:62, wherein said polypeptide comprises an immunogenic portion of a Leishmania antigen, said immunogenic portion of Leishmania antigen selectively binds to anti-leishmania antibodies.

9. An isolated polypeptide comprising SEQ ID NO:80.

10. An isolated polypeptide comprising SEQ ID NO:81.

11. An isolated polypeptide comprising SEQ ID NO:83.

12. An isolated polypeptide comprising SEQ ID NO:84.

13. An isolated polypeptide comprising SEQ ID NO:85.

14. An isolated polypeptide comprising a sequence selected from the group consisting of SEQ ID NO: 86 and 87.

[Previous Doc](#)

[Next Doc](#)

[Go to Doc#](#)

[First Hit](#) [Fwd Refs](#)[Previous Doc](#)[Next Doc](#)[Go to Doc#](#)

Generate Collection

Print

L15: Entry 43 of 71

File: USPT

Dec 31, 2002

DOCUMENT-IDENTIFIER: ~~US 6500437-B1~~TITLE: Leishmania antigens for use in the therapy and diagnosis of leishmaniasisAbstract Text (1):

Compositions and methods for preventing, treating and detecting leishmaniasis and stimulating immune responses in patients are disclosed. The compounds provided include polypeptides that contain at least an immunogenic portion of one or more Leishmania antigens, or a variant thereof. Vaccines and pharmaceutical compositions comprising such polypeptides, or polynucleotides encoding such polypeptides, are also provided and may be used, for example, for the prevention and therapy of leishmaniasis, as well as for the detection of Leishmania infection.

Brief Summary Text (2):

The present invention relates generally to compositions and methods for preventing, treating and detecting leishmaniasis, and for stimulating immune responses in patients. The invention is more particularly related to polypeptides comprising an immunogenic portion of a Leishmania antigen or a variant thereof, and to vaccines and pharmaceutical compositions comprising one or more such polypeptides. The vaccines and pharmaceutical compositions may be used, for example, for the prevention and therapy of leishmaniasis, as well as for the detection of Leishmania infection.

Brief Summary Text (4):

Leishmania organisms are intracellular protozoan parasites of macrophages that cause a wide range of clinical diseases in humans and domestic animals, primarily dogs. In some infections, the parasite may lie dormant for many years. In other cases, the host may develop one of a variety of forms of leishmaniasis. For example, the disease may be asymptomatic or may be manifested as subclinical visceral leishmaniasis, which is characterized by mild symptoms of malaise, diarrhea and intermittent hepatomegaly. Patients with subclinical or asymptomatic disease usually have low antibody titers, making the disease difficult to detect with standard techniques. Alternatively, leishmaniasis may be manifested as a cutaneous disease, which is a severe medical problem but is generally self-limiting, or as a highly destructive mucosal disease, which is not self-limiting. Finally, and most seriously, the disease may be manifested as an acute visceral infection involving the spleen, liver and lymph nodes, which, untreated, is generally a fatal disease. Symptoms of acute visceral leishmaniasis include hepatosplenomegaly, fever, leukopenia, anemia and hypergammaglobulinemia.

Brief Summary Text (6):

Accurate diagnosis of leishmaniasis is frequently difficult to achieve. There are 20 species of Leishmania that infect humans, including L. donovani, L. chagasi, L. infantum, L. major, L. amazonensis, L. braziliensis, L. panamensis, L. mexicana, L. tropica, and L. guyanensis, and there are no distinctive signs or symptoms that unambiguously indicate the presence of Leishmania infection. Parasite detection methods have been used, but such methods are neither sensitive nor clinically practical. Current skin tests typically use whole or lysed parasites. Such tests are generally insensitive, irreproducible and prone to cross-reaction with a variety of other diseases. In addition, the preparations employed in such tests are often unstable. Thus, there is a need for improved methods for the detection of Leishmania infection.

Brief Summary Text (9):

h e b b g e e f c e

e ge

Briefly stated, the present invention provides compositions and methods for preventing, treating and detecting leishmaniasis, as well as for stimulating immune responses in patients. In one aspect, polypeptides are provided which comprise at least an immunogenic portion of a Leishmania antigen, or a variant of such an antigen that differs only in conservative substitutions and/or modifications. In specific embodiments of the invention, the Leishmania antigen comprises an amino acid sequence selected from the group consisting of SEQ ID NO: 2, 4, 20, 22, 24, 26, 36-38, 41, 50-53 and 82. DNA sequences encoding the above polypeptides, recombinant expression vectors comprising these DNA sequences and host cells transformed or transfected with such expression vectors are also provided.

Brief Summary Text (10):

In further aspects, the present invention provides fusion proteins comprising Leishmania antigens, together with polynucleotides encoding such fusion proteins. In certain specific embodiments, such fusion proteins comprise an amino acid sequence selected from the group consisting of SEQ ID NO: 95, 96 and 97.

Brief Summary Text (11):

In related aspects, the present invention provides pharmaceutical compositions which comprise one or more of the polypeptides and/or fusion proteins described herein, or a polynucleotide encoding such polypeptides and fusion proteins, and a physiologically acceptable carrier. Vaccines which comprise one or more such polypeptides, fusion proteins or polynucleotides, together with an immunostimulant are also provided. In specific embodiments of these aspects, the Leishmania antigen has an amino acid sequence selected from the group consisting of SEQ ID NO: 2, 4, 20, 22, 24, 26, 36-38, 41, 50-53 and 82.

Brief Summary Text (12):

In still further related embodiments, the pharmaceutical compositions and vaccines comprise at least two different polypeptides, each polypeptide comprising an immunogenic portion of a Leishmania antigen having an amino acid sequence selected from the group consisting of sequences recited in SEQ ID NO: 2, 4, 6, 8, 10, 20, 22, 24, 26, 36-38, 41, 50-53, 82, and variants thereof that differ only in conservative substitutions and/or modifications. In other embodiments, the inventive pharmaceutical compositions comprise one or more of the inventive polypeptides in combination with a known Leishmania antigen.

Brief Summary Text (13):

In yet other related embodiments, the pharmaceutical compositions and vaccines comprise soluble Leishmania antigens.

Brief Summary Text (15):

In further aspects, methods and diagnostic kits are provided for detecting Leishmania infection in a patient. The methods comprise: (a) contacting dermal cells of a patient with a pharmaceutical composition as described above; and (b) detecting an immune response on the patient's skin, therefrom detecting Leishmania infection in the patient. The diagnostic kits comprise: (a) a pharmaceutical composition as described above; and (b) an apparatus sufficient to contact the pharmaceutical composition with the dermal cells of a patient.

Drawing Description Text (4):

FIG. 3 illustrates the expression and purification of the Leishmania antigen Ldp23 as a recombinant fusion protein. Panel A shows a Coomassie blue-stained SDS-PAGE gel of lysed E. coli without (lane 1) and with (lane 2) IPTG induction of Ldp23 expression. Arrow indicates the recombinant fusion protein. Panel B shows the fusion protein following excision from a preparative SDS-PAGE gel, electroelution, dialysis against PBS and analytical SDS-PAGE.

Drawing Description Text (8):

FIG. 7 shows the stimulation of Leishmania-specific T-cell proliferation by Ldp23. The results are presented as relative cell number as a function of fluorescence intensity. T-cells (10^{sup}.5 /well) were purified from lymph nodes of BALB/c mice immunized in the foot pad with L. donovani promastigotes in CFA and were cultured with various concentrations of the purified recombinant Ldp23 in the presence of

2.times.10.sup.5 Mitomycin C-treated normal BALB/c spleen mononuclear cells. Proliferation of T-cells was measured at 27 hours of culture. Values are expressed as cpm and represent the mean of [.sup.3 H]TdR incorporation of triplicate cultures.

Drawing Description Text (9):

FIG. 8 illustrates Ldp23-induced cytokine production by lymph node cells of BALB/c mice. Cultures were incubated with varying amounts of Ldp23 or Leishmania lysate, presented as .mu.g/mL, and were assayed by ELISA for the production of interferon-.gamma. (panel A) or interleukin-4 (panel B), both of which are shown as ng/mL.

Drawing Description Text (10):

FIG. 9 shows the PCR amplification of cytokine mRNAs isolated from mucosal leishmaniasis (Panel A) and cutaneous leishmaniasis (panel B) patient PBMC before and after stimulation with representative polypeptides of the present invention. Lanes 0 and--indicate the level of PCR products at the initiation of culture and after 72 hours of culture, respectively, in the absence of added polypeptide; lanes Lb, 83a and 83b indicate the level of PCR products following culturing of PBMC with L. braziliensis lysate, and the Leishmania antigens Lbhsp83a and Lbhsp83b, respectively.

Drawing Description Text (11):

FIG. 10 presents a comparison of the levels of interferon-.gamma. (panel A) and TNF-.alpha. (panel B) in the supernatants of 72 hour PBMC cultures from Leishmania-infected and control individuals in response to stimulation with parasite lysate or the indicated polypeptides.

Drawing Description Text (12):

FIG. 11 illustrates the levels of IL-10p40 (in pg/mL) in the supernatant of PBMC cultures from L. braziliensis-infected individuals and uninfected controls 72 hours following stimulation with parasite promastigote lysate (Lb), Lbhsp83a or Lbhsp83b.

Drawing Description Text (15):

FIG. 14 shows the level of IFN-.gamma. (in pg/mL) secreted by Leishmania-infected and uninfected human PBMC stimulated by the Leishmania antigen M15, as compared to the levels stimulated by L. major lysate and L-Rack, an antigen that does not appear to be recognized by Leishmania-infected humans.

Drawing Description Text (16):

FIG. 15 shows the level of IFN-.gamma. (in pg/mL) secreted by infected and uninfected human PBMC stimulated by soluble Leishmania antigens (S antigens), as compared to the levels stimulated by L. major lysate and L-Rack.

Drawing Description Text (18):

FIG. 17 shows the proliferation of human PBMC, prepared from Leishmania-immune and uninfected individuals, stimulated by M15 as compared to the proliferation stimulated by L. major lysate and L-Rack. Values are expressed as cpm.

Drawing Description Text (19):

FIG. 18 illustrates the proliferation of human PBMC, prepared from Leishmania-infected and uninfected individuals, stimulated by soluble Leishmania antigens as compared to the proliferation stimulated by culture medium, L. major lysate and L-Rack. Values are expressed as cpm.

Drawing Description Text (21):

FIG. 20 illustrates the reactivity of rabbit sera raised against soluble Leishmania antigens with Leishmania promastigote lysate (lane 1) and soluble Leishmania antigens (lane 2).

Drawing Description Text (22):

FIG. 21 shows the cDNA and predicted amino acid sequence for the Leishmania antigen Lmspla.

Drawing Description Text (23):

FIG. 22 shows a Southern blot of genomic DNA from *L. major* digested with a panel of restriction enzymes (lanes 1 to 7) and six other *Leishmania* species digested with PstI (lanes 8 to 13) probed with the full-length cDNA insert of *Lmspla*.

Drawing Description Text (24):

FIG. 23 shows a Southern blot of genomic DNA from *L. major* digested with a panel of restriction enzymes, six other *Leishmania* species digested with PstI and the infectious pathogens *T. cruzi* and *T. brucei*, probed with the full-length cDNA insert of the *Leishmania* antigen MAPS-1A.

Drawing Description Text (29):

FIG. 28 illustrates the effectiveness of immunization with either soluble *Leishmania* antigens or a mixture of Ldp23, LbeiF4A and M15 plus adjuvant in conferring protection against infection (as measured by footpad swelling) in a murine leishmaniasis model system, as compared to the administration of adjuvant alone.

Drawing Description Text (32):

FIG. 31 illustrates the effectiveness of immunization with soluble *Leishmania* antigens, MAPS-1A and M15 plus adjuvant, IL-12, in conferring protection against infection (as measured by footpad swelling) in a murine leishmaniasis model system, as compared to the administration of adjuvant IL-12 alone.

Drawing Description Text (34):

FIG. 33 illustrates the effectiveness of immunization with *Leishmania* fusion proteins plus IL-12 as adjuvant, in conferring protection against infection in a murine leishmaniasis model system.

Drawing Description Text (35):

FIG. 34 illustrates the effectiveness of immunization with *Leishmania* fusion proteins plus the adjuvant MPL-SE, in conferring protection against infection in a murine leishmaniasis model system.

Detailed Description Text (2):

As noted above, the present invention is generally directed to compositions and methods for preventing, treating and detecting leishmaniasis, as well as for stimulating immune responses in patients. The compositions of the subject invention include polypeptides that comprise at least an immunogenic portion of a *Leishmania* antigen, or a variant of such an antigen that differs only in conservative substitutions and/or modifications. In one preferred embodiment, compositions of the present invention include multiple polypeptides selected so as to provide enhanced protection against a variety of *Leishmania* species.

Detailed Description Text (3):

Polypeptides within the scope of the present invention include, but are not limited to, polypeptides comprising immunogenic portions of *Leishmania* antigens comprising the sequences recited in SEQ ID NO:2 (referred to herein as M15), SEQ ID NO:4 (referred to herein as Ldp23), SEQ ID NO:6 (referred to herein as Lbhsp83), SEQ ID NO:8 (referred to herein as Lt-210), SEQ ID NO:10 (referred to herein as LbeiF4A), SEQ ID NO: 20 (referred to herein as *Lmspla*), SEQ ID NO: 22 (referred to herein as *Lmsp9a*), SEQ ID NOs: 24 and 26 (referred to herein as MAPS-1A), and SEQ ID NO: 36-42, 49-53 and 55. As used herein, the term "polypeptide" encompasses amino acid chains of any length, including full length proteins (i.e., antigens), wherein the amino acid residues are linked by covalent bonds. Thus, a polypeptide comprising an immunogenic portion of one of the above antigens may consist entirely of the immunogenic portion, or may contain additional sequences. The additional sequences may be derived from the native *Leishmania* antigen or may be heterologous, and such sequences may (but need not) be immunogenic. An antigen "having" a particular sequence is an antigen that contains, within its full length sequence, the recited sequence. The native antigen may, or may not, contain additional amino acid sequence.

Detailed Description Text (4):

h e b b g e e f c e e ge

An immunogenic portion of a Leishmania antigen is a portion that is capable of eliciting an immune response (i.e., cellular and/or humoral) in a presently or previously Leishmania-infected patient (such as a human or a dog) and/or in cultures of lymph node cells or peripheral blood mononuclear cells (PBMC) isolated from presently or previously Leishmania-infected individuals. The cells in which a response is elicited may comprise a mixture of cell types or may contain isolated component cells (including, but not limited to, T-cells, NK cells, macrophages, monocytes and/or B cells). In particular, immunogenic portions are capable of inducing T-cell proliferation and/or a dominantly Th1-type cytokine response (e.g., IL-2, IFN- γ , and/or TNF- α production by T-cells and/or NK cells; and/or IL-12 production by monocytes, macrophages and/or B cells). Immunogenic portions of the antigens described herein may generally be identified using techniques known to those of ordinary skill in the art, including the representative methods provided herein.

Detailed Description Text (9):

"Polypeptides" as described herein also include combination polypeptides, also referred to as fusion proteins. A "combination polypeptide" is a polypeptide comprising at least one of the above immunogenic portions and one or more additional immunogenic Leishmania sequences, which are joined via a peptide linkage into a single amino acid chain. The sequences may be joined directly (i.e., with no intervening amino acids) or may be joined by way of a linker sequence (e.g., Gly-Cys-Gly) that does not significantly diminish the immunogenic properties of the component polypeptides.

Detailed Description Text (12):

The ligated DNA sequences are operably linked to suitable transcriptional or translational regulatory elements. The regulatory elements responsible for expression of DNA are located only 5' to the DNA sequence encoding the first polypeptides. Similarly, stop codons required to end translation and transcription termination signals are only present 3' to the DNA sequence encoding the second polypeptide. The preparation of fusion proteins of Leishmania antigens is described below in Example 19.

Detailed Description Text (13):

In general, Leishmania antigens having immunogenic properties, and DNA sequences encoding such antigens, may be prepared using any of a variety of procedures from one or more Leishmania species including, but not limited to, L. donovani, L. chagasi, L. infantum, L. major, L. amazonensis, L. braziliensis, L. panamensis, L. mexicana, L. tropica, and L. guyanensis. Such species are available, for example, from the American Type Culture Collection (ATCC), Rockville, Md. For example, peptides isolated from MHC class II molecules of macrophages infected with a Leishmania species may be used to rescue the corresponding Leishmania donor antigens. MHC class II molecules are expressed mainly by cells of the immune system, including macrophages. These molecules present peptides, which are usually 13-17 amino acids long, derived from foreign antigens that are degraded in cellular vesicles. The bound peptide antigens are then recognized by CD4 T-cells. Accordingly, foreign peptides isolated from MHC class II molecules of, for example, Leishmania-infected murine macrophages may be used to identify immunogenic Leishmania proteins.

Detailed Description Text (14):

Briefly, peptides derived from Leishmania antigens may be isolated by comparing the reverse phase HPLC profile of peptides extracted from infected macrophages with the profile of peptides extracted from uninfected cells. Peptides giving rise to distinct HPLC peaks unique to infected macrophages may then be sequenced using, for example, Edman chemistry as described in Edman and Berg, Eur J. Biochem, 80:116-132 (1967). A DNA fragment corresponding to a portion of a Leishmania gene encoding the peptide may then be amplified from a Leishmania cDNA library using an oligonucleotide sense primer derived from the peptide sequence and an oligo dT antisense primer. The resulting DNA fragment may then be used as a probe to screen a Leishmania library for a full length cDNA or genomic clone that encodes the Leishmania antigen. Such screens may generally be performed using techniques well known to those of ordinary skill in the art, such as those described in Sambrook et

al., Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratories, Cold Spring Harbor, N.Y. (1989).

Detailed Description Text (15):

This approach may be used to identify a 23 kD Leishmania donovani antigen (referred to herein as Ldp23). The sequence of a polynucleotide encoding Ldp23 is provided in SEQ ID NO:3 and the amino acid sequence of Ldp23 is provided in SEQ ID NO:4. Using the methods described herein, Ldp23 has been shown to induce a Th1 immune response in T-cells prepared from Leishmania-infected mice.

Detailed Description Text (16):

Alternatively, a Leishmania cDNA or genomic expression library may be screened with serum from a Leishmania-infected individual, using techniques well known to those of ordinary skill in the art. Polynucleotides encoding reactive antigens may then be used to express the recombinant antigen for purification. The immunogenic properties of the purified Leishmania antigens may then be evaluated using, for example the representative methods described herein.

Detailed Description Text (17):

For example, sera from Leishmania-infected mice may be used to screen a cDNA library prepared from Leishmania amastigotes. Reactive clones may then be expressed and recombinant proteins assayed for the ability to stimulate T-cells or NK cells derived from Leishmania-immune individuals (i.e., individuals having evidence of infection, as documented by positive serological reactivity with Leishmania-specific antibodies and/or a Leishmania-specific DTH response, without clinical symptoms of leishmaniasis). This procedure may be used to obtain a recombinant polynucleotide encoding the Leishmania antigen designated M15. The sequence of such a polynucleotide is provided in SEQ ID NO:1, and the amino acid sequence of the encoded protein is provided in SEQ ID NO:2.

Detailed Description Text (18):

A similar approach may be used to isolate a genomic polynucleotide encoding an immunogenic Leishmania braziliensis antigen, referred to herein as Lbhsp83. More specifically, a genomic clone encoding Lbhsp83 may be isolated by screening a L. braziliensis expression library with sera from a Leishmania-infected individual. The DNA encoding Lbhsp83 is homologous to the gene encoding the eukaryotic 83 kD heat shock protein. The sequence of a polynucleotide encoding nearly all of Lbhsp83 is presented in SEQ ID NO:5, and the encoded amino acid sequence is provided in SEQ ID NO:6. Using the methods described below, Lbhsp83 has been found to stimulate proliferation, and a mixed Th1 and Th2 cytokine profile, in PBMC isolated from L. braziliensis-infected patients. Accordingly, Lbhsp83 is an immunogenic Leishmania antigen. Regions of Lbhsp83 that are not conserved with the mammalian gene have been found to be particularly potent for T-cell stimulation and antibody binding. Such regions may be identified, for example, by visual inspection of the sequence comparison provided in FIG. 19.

Detailed Description Text (20):

The above approach may further be used to isolate a polynucleotide encoding a L. braziliensis antigen referred to herein as LbeIF4A. Briefly, such a clone may be isolated by screening a L. braziliensis expression library with sera obtained from a patient afflicted with mucosal leishmaniasis, and analyzing the reactive antigens for the ability to stimulate proliferative responses and preferential Th1 cytokine production in PBMC isolated from Leishmania-infected patients, as described below. The preparation and characterization of LbeIF4A is described in detail in U.S. patent application Ser. Nos. 08/454,036 and 08/488,386, which are continuations-in-part of U.S. patent application Ser. No. 08/232,534, filed Apr. 22, 1994. The sequence of a polynucleotide encoding LbeIF4A is provided in SEQ ID NO:9 and the encoded amino acid sequence is presented in SEQ ID NO:10. Homologs of LbeIF4A, such as that found in L. major, may also be isolated using this approach, and are within the scope of the present invention.

Detailed Description Text (21):

Compositions of the present invention may also, or alternatively, contain soluble Leishmania antigens. As used herein, "soluble Leishmania antigens" refers to a

mixture of at least 8 different Leishmania antigens that may be isolated from the supernatant of Leishmania promastigotes of any species grown for 8-12 hours in protein-free medium. Briefly, the organisms are grown to late log phase in complex medium with serum until they reach a density of 2-3.times.10.sup.7 viable organisms per mL of medium. The organisms are thoroughly washed to remove medium components and resuspended at 2-3.times.10.sup.7 viable organisms per mL of defined serum-free medium consisting of equal parts RPMI 1640 and medium 199, both from Gibco BRL, Gaithersburg, Md. After 8-12 hours. the supernatant containing soluble Leishmania antigens is removed, concentrated 10 fold and dialyzed against phosphate-buffered saline for 24 hours. The presence of at least eight different antigens within the mixture of Leishmania antigens may be confirmed using SDS-PAGE (i.e., through the observation of at least 8 different bands). The immunogenic properties of the soluble Leishmania antigens may be confirmed by evaluating the ability of the preparation to elicit an immune response in cultures of lymph node cells and/or peripheral blood mononuclear cells (PBMC) isolated from presently or previously Leishmania-infected individuals. Such an evaluation may be performed as described below.

Detailed Description Text (22):

Individual antigens present within the mixture of soluble Leishmania antigens may be isolated by immunizing mice or rabbits with Leishmania culture supernatant, containing soluble antigens, and employing the resultant sera to screen a Leishmania cDNA expression library as described in detail below. This procedure may be used to isolate recombinant polynucleotides encoding the L. major antigens referred to herein as Lmspla, Lmsp9a and MAPS-1A. DNA sequences encoding Lmspla, Lmsp9a and MAPS-1A are provided in SEQ ID NO: 19, 21 and 23, respectively, with the corresponding predicted amino acid sequences being presented in SEQ ID NO: 20, 22 and 24, respectively. Similarly, sera from mice or rabbits immunized with L. major culture supernatant may be used to screen an L. major genomic DNA library. As detailed below, this procedure may be used to isolate polynucleotides encoding the L. major antigens referred to herein as LmgSP1, LmgSP3, LmgSP5, LmgSP8, LmgSP9, LmgSP13, LmgSP19, and polynucleotides encoding the L. chagasi antigens LcgSP1, LcgSP3, LcgSP4, LcgSP8, and LcgSP10. The DNA sequences encoding these antigens are provided in SEQ ID NO:29-35 and 44-48, respectively, with the corresponding amino acid sequences being provided in SEQ ID NO: 36-42 and 49-53. The L. major antigens referred to herein as 1G6-34, 1E6-44, 4A5-63, 1B11-39, 2A10-37, 4G2-83, 4H6-41 and 8G3-100 may be isolated by means of CD4+ T cell expression cloning as described below. DNA sequences encoding these antigens are provided in SEQ ID NO: 72-79, respectively, with the corresponding predicted amino acid sequences being provided in SEQ ID NO: 80-87. The immunogenic properties of the isolated Leishmania antigens may be evaluated using, for example, the representative methods described herein.

Detailed Description Text (23):

Regardless of the method of preparation, the antigens described herein are immunogenic. In other words, the antigens (and immunogenic portions thereof) are capable of eliciting an immune response in cultures of lymph node cells and/or peripheral blood mononuclear cells (PBMC) isolated from presently or previously Leishmania-infected individuals. More specifically, the antigens, and immunogenic portions thereof, have the ability to induce T-cell proliferation and/or to elicit a dominantly Th1-type cytokine response (e.g., IL-2, IFN-.gamma., and/or TNF-.alpha. production by T-cells and/or NK cells; and/or IL-12 production by monocytes, macrophages and/or B cells) in cells isolated from presently or previously Leishmania-infected individuals. A Leishmania-infected individual may be afflicted with a form of leishmaniasis (such as subclinical, cutaneous, mucosal or active visceral) or may be asymptomatic. Such individuals may be identified using methods known to those of ordinary skill in the art. Individuals with leishmaniasis may be identified based on clinical findings associated with at least one of the following: isolation of parasite from lesions, a positive skin test with Leishmania lysate or a positive serological test. Asymptomatic individuals are infected individuals who have no signs or symptoms of the disease. Such individuals can be identified based on a positive serological test and/or skin test with Leishmania lysate.

Detailed Description Text (24):

h e b b g e e f c e e ge

The term "PBMC," which refers to a preparation of nucleated cells consisting primarily of lymphocytes and monocytes that are present in peripheral blood, encompasses both mixtures of cells and preparations of one or more purified cell types. PBMC may be isolated by methods known to those in the art. For example, PBMC may be isolated by density centrifugation through, for example, Ficoll.TM.

(Winthrop Laboratories, New York). Lymph node cultures may generally be prepared by immunizing BALB/c mice (e.g., in the rear foot pad) with Leishmania promastigotes emulsified in complete Freund's adjuvant. The draining lymph nodes may be excised following immunization and T-cells may be purified in an anti-mouse Ig column to remove the B cells, followed by a passage through a Sephadex G10 column to remove the macrophages. Similarly, lymph node cells may be isolated from a human following biopsy or surgical removal of a lymph node.

Detailed Description Text (25):

The ability of a polypeptide (e.g., a Leishmania antigen or a portion or other variant thereof) to induce a response in PBMC or lymph node cell cultures may be evaluated by contacting the cells with the polypeptide and measuring a suitable response. In general, the amount of polypeptide that is sufficient for the evaluation of about 2.times.10.sup.5 cells ranges from about 10 ng to about 100 .mu.g, and preferably is about 1-10 .mu.g. The incubation of polypeptide with cells is typically performed at 37.degree. C. for about 1-3 days. Following incubation with polypeptide, the cells are assayed for an appropriate response. If the response is a proliferative response, any of a variety of techniques well known to those of ordinary skill in the art may be employed. For example, the cells may be exposed to a pulse of radioactive thymidine and the incorporation of label into cellular DNA measured. In general, a polypeptide that results in at least a three fold increase in proliferation above background (i.e., the proliferation observed for cells cultured without polypeptide) is considered to be able to induce proliferation.

Detailed Description Text (26):

Alternatively, the response to be measured may be the secretion of one or more cytokines (such as interferon-.gamma. (IFN-.gamma.), interleukin-4 (IL-4), interleukin-12 (p70 and/or p40), interleukin-2 (IL-2) and/or tumor necrosis factor-.alpha. (TNF-.alpha.)) or the change in the level of mRNA encoding one or more specific cytokines. In particular, the secretion of interferon-.gamma., interleukin-2, tumor necrosis factor-.alpha. and/or interleukin-12 is indicative of a Th1 response, which is responsible for the protective effect against Leishmania. Assays for any of the above cytokines may generally be performed using methods known to those of ordinary skill in the art, such as an enzyme-linked immunosorbent assay (ELISA). Suitable antibodies for use in such assays may be obtained from a variety of sources such as Chemicon, Temucula, Calif. and PharMingen, San Diego, Calif., and may generally be used according to the manufacturer's instructions. The level of mRNA encoding one or more specific cytokines may be evaluated by, for example, amplification by polymerase chain reaction (PCR). In general, a polypeptide that is able to induce, in a preparation of about 1-3.times.10.sup.5 cells, the production of 30 pg/mL of IL-12, IL-4, IFN-.gamma., TNF-.alpha. or IL-12 p40, or 10 pg/mL of IL-12 p70, is considered able to stimulate production of a cytokine.

Detailed Description Text (28):

Portions and other variants of immunogenic Leishmania antigens may be generated by synthetic or recombinant means. Synthetic polypeptides having fewer than about 100 amino acids, and generally fewer than about 50 amino acids, may be generated using techniques well known to those of ordinary skill in the art. For example, such polypeptides may be synthesized using any of the commercially available solid-phase techniques, such as the Merrifield solid-phase synthesis method, where amino acids are sequentially added to a growing amino acid chain. See Merrifield, J. Am. Chem. Soc. 85:2149-2146, 1963. Equipment for automated synthesis of polypeptides is commercially available from suppliers such as Perkin Elmer/Applied BioSystems Division, Foster City, Calif., and may be operated according to the manufacturer's instructions.

Detailed Description Text (31):

h e b b g e e f c e e ge

In another aspect, the present invention provides epitope repeat sequences, or antigenic epitopes, of a Leishmania antigen, together with polypeptides comprising at least two such contiguous antigenic epitopes. As used herein an "epitope" is a portion of an antigen that reacts with sera from Leishmania-infected individuals (i.e. an epitope is specifically bound by one or more antibodies present in such sera). As discussed above, epitopes of the antigens described in the present application may be generally identified using techniques well known to those of skill in the art.

Detailed Description Text (32):

In one embodiment, antigenic epitopes of the present invention comprise an amino acid sequence provided in SEQ ID NO:43, 56, 57 or 58. As discussed in more detail below, antigenic epitopes provided herein may be employed in the diagnosis and treatment of Leishmania infection, either alone or in combination with other Leishmania antigens or antigenic epitopes. Antigenic epitopes and polypeptides comprising such epitopes may be prepared by synthetic means, as described generally above and in detail in Example 15.

Detailed Description Text (33):

In certain aspects of the present invention, described in detail below, the polypeptides, antigenic epitopes and/or soluble Leishmania antigens may be incorporated into pharmaceutical compositions or vaccines. For clarity, the term "polypeptide" will be used when describing specific embodiments of the inventive therapeutic compositions and diagnostic methods. However, it will be clear to one of skill in the art that the antigenic epitopes of the present invention may also be employed in such compositions and methods. Pharmaceutical compositions comprise one or more polypeptides, each of which may contain one or more of the above sequences (or variants thereof), and a physiologically acceptable carrier. Vaccines comprise one or more of the above polypeptides and an immunostimulant, such as an adjuvant (e.g., IbeIF4A, interleukin-12 or other cytokines) or a liposome (into which the polypeptide is incorporated). In certain embodiments, the inventive vaccines include an adjuvant capable of eliciting a predominantly Th-1 type response. Preferred adjuvants for use in eliciting a predominantly Th1-type response include, for example, a combination of monophosphoryl lipid A, preferably 3-de-O-acylated monophosphoryl lipid A (3D-MPL), together with an aluminum salt. MPL adjuvants are available from Ribi ImmunoChem Research Inc. (Hamilton, Mont.; see U.S. Pat. Nos. 4,436,727; 4,877,611; 4,866,034 and 4,912,094). CpG-containing oligonucleotides (in which the CpG dinucleotide is unmethylated) also induce a predominantly Th1 response. Such oligonucleotides are well known and are described, for example, in WO 96/02555 and WP 99/33488. Immunostimulatory DNA sequences are also described, for example, by Sato et al., Science 273:352, 1996. Another preferred adjuvant is a saponin, preferably QS21 (Aquila, United States), which may be used alone or in combination with other adjuvants. For example, an enhanced system involves the combination of a monophosphoryl lipid A and saponin derivative, such as the combination of QS21 and 3D-MPL as described in WO 94/00153, or a less reactogenic composition where the QS21 is quenched with cholesterol, as described in WO 96/33739. Other preferred formulations comprise an oil-in-water emulsion and tocopherol. A particularly potent adjuvant formulation involving QS21, 3D-MPL and tocopherol in an oil-in-water emulsion is described in WO 95/17210.

Detailed Description Text (34):

Vaccines may additionally contain a delivery vehicle, such as a biodegradable microsphere (disclosed, for example, in U.S. Pat. Nos. 4,897,268 and 5,075,109). Pharmaceutical compositions and vaccines within the scope of the present invention may also contain other Leishmania antigens, either incorporated into a combination polypeptide or present within one or more separate polypeptides.

Detailed Description Text (38):

In one preferred embodiment, compositions of the present invention include multiple polypeptides selected so as to provide enhanced protection against a variety of Leishmania species. Such polypeptides may be selected based on the species of origin of the native antigen or based on a high degree of conservation of amino acid sequence among different species of Leishmania. A combination of individual polypeptides may be particularly effective as a prophylactic and/or therapeutic

vaccine because (1) stimulation of proliferation and/or cytokine production by a combination of individual polypeptides may be additive, (2) stimulation of proliferation and/or cytokine production by a combination of individual polypeptides may be synergistic, (3) a combination of individual polypeptides may stimulate cytokine profiles in such a way as to be complementary to each other and/or (4) individual polypeptides may be complementary to one another when certain of them are expressed more abundantly on the individual species or strain of Leishmania responsible for infection. A preferred combination contains polypeptides that comprise immunogenic portions of M15, Ldp23, Lbhsp83, Lt-1 and LbeIF4A. Alternatively, or in addition, the combination may include one or more polypeptides comprising immunogenic portions of other Leishmania antigens disclosed herein, and/or soluble Leishmania antigens.

Detailed Description Text (39):

In another preferred embodiment, compositions of the present invention include single polypeptides selected so as to provide enhanced protection against a variety of Leishmania species. A single individual polypeptide may be particularly effective as a prophylactic and/or therapeutic vaccine for those reasons stated above for combinations of individual polypeptides.

Detailed Description Text (40):

In another embodiment, compositions of the present invention include individual polypeptides and combinations of the above described polypeptides employed with a variety of adjuvants, such as IL-12 (protein or DNA) to confer a protective response against a variety of Leishmania species.

Detailed Description Text (41):

In yet another embodiment, compositions of the present invention include DNA constructs of the various Leishmania species employed alone or in combination with variety of adjuvants, such as IL-12 (protein or DNA) to confer a protective response against a variety of Leishmania species.

Detailed Description Text (42):

The above pharmaceutical compositions and vaccines may be used, for example, to induce protective immunity against Leishmania in a patient, such as a human or a dog, to prevent leishmaniasis. Appropriate doses and methods of administration for this purposes are described in detail below.

Detailed Description Text (43):

The pharmaceutical compositions and vaccines described herein may also be used to stimulate an immune response, which may be cellular and/or humoral, in a patient. For Leishmania-infected patients, the immune responses that may be generated include a preferential Th1 immune response (i.e., a response characterized by the production of the cytokines interleukin-1, interleukin-2, interleukin-12 and/or interferon-.gamma., as well as tumor necrosis factor-.alpha.). For uninfected patients, the immune response may be the production of interleukin-12 and/or interleukin-2, or the stimulation of gamma delta T-cells. In either category of patient, the response stimulated may include IL-12 production. Such responses may also be elicited in biological samples of PBMC or components thereof derived from Leishmania-infected or uninfected individuals. As noted above, assays for any of the above cytokines may generally be performed using methods known to those of ordinary skill in the art, such as an enzyme-linked immunosorbent assay (ELISA).

Detailed Description Text (44):

Suitable pharmaceutical compositions and vaccines for use in this aspect of the present invention are those that contain at least one polypeptide comprising an immunogenic portion of a Leishmania antigen disclosed herein (or a variant thereof). Preferably, the polypeptides employed in the pharmaceutical compositions and vaccines are complementary, as described above. Soluble Leishmania antigens, with or without additional polypeptides, may also be employed.

Detailed Description Text (45):

The pharmaceutical compositions and vaccines described herein may also be used to treat a patient afflicted with a disease responsive to IL-12 stimulation. The

patient may be any warm-blooded animal, such as a human or a dog. Such diseases include infections (which may be, for example, bacterial, viral or protozoan) or diseases such as cancer. In one embodiment, the disease is leishmaniasis, and the patient may display clinical symptoms or may be asymptomatic. In general, the responsiveness of a particular disease to IL-12 stimulation may be determined by evaluating the effect of treatment with a pharmaceutical composition or vaccine of the present invention on clinical correlates of immunity. For example, if treatment results in a heightened Th1 response or the conversion of a Th2 to a Th1 profile, with accompanying clinical improvement in the treated patient, the disease is responsive to IL-12 stimulation. Polypeptide administration may be as described below, or may extend for a longer period of time, depending on the indication. Preferably, the polypeptides employed in the pharmaceutical compositions and vaccines are complementary, as described above. A particularly preferred combination contains polypeptides that comprise immunogenic portions of M15, Ldp23, Lbhsp83, Lt-1 and LbeIF4A, Lmspla, Lmsp9a, and MAPS-1A. Soluble Leishmania antigens, with or without additional polypeptides, may also be employed.

Detailed Description Text (47):

In another aspect, this invention provides methods for using one or more of the polypeptides described above to diagnose Leishmania infection in a patient using a skin test. As used herein, a "skin test" is any assay performed directly on a patient in which a delayed-type hypersensitivity (DTH) reaction (such as induration and accompanying redness) is measured following intradermal injection of one or more polypeptides as described above. Such injection may be achieved using any suitable device sufficient to contact the polypeptide or polypeptides with dermal cells of the patient, such as a tuberculin syringe or 1 mL syringe. Preferably, the reaction is measured at least 48 hours after injection, more preferably 72 hours after injection.

Detailed Description Text (48):

The DTH reaction is a cell-mediated immune response, which is greater in patients that have been exposed previously to a test antigen (i.e., an immunogenic portion of a polypeptide employed, or a variant thereof). The response may be measured visually, using a ruler. In general, induration that is greater than about 0.5 cm in diameter, preferably greater than about 1.0 cm in diameter, is a positive response, indicative of Leishmania infection, which may or may not be manifested as an active disease.

Detailed Description Text (49):

The polypeptides of this invention are preferably formulated, for use in a skin test, as pharmaceutical compositions containing at least one polypeptide and a physiologically acceptable carrier, as described above. Such compositions typically contain one or more of the above polypeptides in an amount ranging from about 1 .mu.g to 100 .mu.g, preferably from about 10 .mu.g to 50 .mu.g in a volume of 0.1 mL. Preferably, the carrier employed in such pharmaceutical compositions is a saline solution with appropriate preservatives, such as phenol and/or Tween 80.TM..

Detailed Description Text (50):

The inventive polypeptides may also be employed in combination with one or more known Leishmania antigens in the diagnosis of leishmaniasis, using, for example, the skin test described above. Preferably, individual polypeptides are chosen in such a way as to be complementary to each other. Examples of known Leishmania antigens which may be usefully employed in conjunction with the inventive polypeptides include K39 (Bums et al., Proc. Natl. Acad. Sci. USA, 1993 90:775-779).

Detailed Description Text (55):

This Example illustrates the preparation of a Leishmania antigen M15, having the sequence provided in SEQ ID NO:2.

Detailed Description Text (58):

The complete insert of clone pfl-1 was excised by digestion with BamHI/KpnI and was subcloned in frame into BamHI/KpnI digested pQE31 (QUIAGEN) to generate the

h e b b g e e f c e

e ge

construct pM151A. *E. coli* containing this construct inducibly expressed high levels of the L. major antigen encoded by pfl1-1 (designated as M15) with the addition of a 6-histidine tag at the amino terminus. Large volume cultures (500 ml) of *E. coli* host cells containing the pM151A construct were induced to express recombinant protein by the addition of 2 mM IPTG at mid-log phase of growth. Growth was continued for 4 to 5 hours and bacteria were then pelleted and washed once with cold PBS. Bacteria were resuspended in 20 ml of lysis buffer (50 mM Na.sub.2 HPO.sub.4, pH 8.0, 300 mM NaCl, 10 mM .beta.-mercaptoethanol) containing 20 mg of lysozyme and were lysed by a 1 hour incubation at 4.degree. C. followed by brief sonication. Insoluble material was removed by centrifugation at 10,000.times.g for 10 minutes and although the recombinant protein was found to be evenly distributed between the soluble and insoluble fractions the insoluble material was discarded at this point. Recombinant protein containing the amino terminal histidine tag was affinity purified using Ni-NTA resin (QIAGEN) according to the manufacturer's recommendations. Briefly, 8 ml of Ni-NTA resin resuspended in lysis buffer was added to the soluble lysate fraction and binding was conducted with constant mixing for 1 hour at 4.degree. C. The mixture was then loaded into a gravity flow column and the non-binding material was allowed to flow through. The Ni-NTA matrix was washed 3 times with 25 ml of wash buffer (50 mM Na.sub.2 HPO.sub.4, pH 6.0, 300 mM NaCl, 10 mM .beta.-mercaptoethanol) and bound material was eluted in 25 ml of elution buffer (50 mM Na.sub.2 HPO.sub.4, pH 5.0, 300 mM NaCl, 10 mM .beta.-mercaptoethanol). The eluted material was then dialyzed against 3 changes of PBS, sterile filtered and stored at -20.degree. C. The purified recombinant protein was shown by SDS-PAGE analysis to be free of any significant amount of *E. coli* protein. A small number of bands of lower molecular weight were assumed to be proteolytic products of the L. major antigen based on their reactivity by western blot analysis. A high titre polyclonal antisera against M15 was generated in rabbits by repeated subcutaneous injection of recombinant protein. Western blot analysis of lysates from L. major promastigotes and amastigotes using this antisera indicated that the protein is constitutively expressed throughout the parasite lifecycle.

Detailed Description Text (61):

This Example illustrates the preparation of a Leishmania antigen Ldp23, having the sequence provided in SEQ ID NO:4.

Detailed Description Text (63):

To ascertain that in vitro infection of macrophages would load their MHC class II molecules with parasite peptides, initial experiments were carried out to test the ability of L. donovani-infected macrophage cell line P388D1 to present parasite antigens to L. donovani specific T-cells. This macrophage cell line was chosen because it has the same H-2 haplotype as the BALB/c mouse, which is a strain of mouse moderately susceptible to L. donovani infection and selected to conduct the in vivo experiments. Using a proportion of 3-5 parasites per cell and an initial incubation at room temperature for 4-6 hours followed by 37.degree. C. for 24-48 hours, close to 90% of the macrophages were infected. The level of MHC class II molecule expression, as determined by FACS analysis, indicated that infection did not cause an effect on the levels of MHC class II expression when compared to non-infected control cells.

Detailed Description Text (64):

To test the ability of the L. donovani-infected P388D1 cells to present parasite antigens, macrophages were infected as indicated above and incubated at 26.degree. C. for 6 hours, and then at 37.degree. C. for either 24, 48 or 72 hours. At each of these time points the non-adherent cells and free parasites were washed out and the adherent cells were mechanically dislodged, washed and fixed with paraformaldehyde. These cells were then used as antigen presenting cells (APCs) for purified lymph node T-cells from BALB/c mice immunized with L. donovani promastigotes. To generate these anti-L. donovani specific T-cells, BALB/c mice (H-2.sup.d) of both sexes (The Jackson Laboratory, Bar Harbor, Me.) were immunized at 8 to 14 weeks of age in the rear foot pad with 5-10.times.10.sup.6 L. donovani promastigotes emulsified in complete Freund's adjuvant (CFA) (Difco Laboratories, Madison, Mich.) as described in Rodrigues et al., Parasite Immunol. 14:49 (1992). The draining lymph nodes were excised 8 days after the immunization and T-cells were purified in an anti-mouse Ig column to remove the B cells, as described in Bunn-Moreno and Campos-Neto, J.

Immunol. 127:427 (1981), followed by a passage through a Sephadex G10 column to remove the macrophages.

Detailed Description Text (69):

Out of three independent peptide extractions, twenty five distinct HPLC peptide peaks were isolated from *L. donovani*-infected macrophages and were subjected to protein sequence analysis using automated Edman degradation on an Applied Biosystems 477 gas-phase protein sequencer. Protein sequence and amino acid analysis were performed by the W.M. Keck Foundation, Biotechnology Resource Laboratory, Yale University, New Haven, Conn. In practically all determinations, no assignment could be made for the first position. Also, in most cases the definition of the amino acid residues of the 10-15 positions was based on the quantitative dominance of one residue over others. Using this approach, the sequences obtained for several peptides showed the presence of 3-6 different residues in many of the 10-15 sequence cycles analyzed for each determination, reflecting a mixture of peptides. In addition, sequences could not be obtained for some peaks because the peptides were blocked. Notwithstanding, three peptides sequences were determined. Amino-acid sequences were searched for identity with proteins in the GenBank database using the GENPETP, PIR and SWISSPROT programs. The sequence data base analysis revealed that one of the peptides was highly homologous to glyceraldehyde-3-phosphate dehydrogenase of various species. Another peptide had homology with elongation factor of several species, including *Leishmania*. The third sequence was not clearly related to any known proteins, and is shown below: XQXPQ(L/K)VFDEXX (SEQ ID NO:11).

Detailed Description Text (72):

The gene fragment was amplified from a *L. donovani* promastigote cDNA preparation using the following reaction conditions: one cycle of 3 min at 94.degree. C. immediately followed by 35 cycles of 1 min at 94.degree. C., 1 min at 45.degree. C. and 1 min at 72.degree. C. The *L. donovani* cDNA was prepared from 5.times.10.sup.7 washed promastigote forms harvested at the log growth phase (3 days culture). The cDNA was obtained using an Invitrogen cDNA cycle.TM. kit (Invitrogen Co., San Diego, Calif.). Oligonucleotide primers were synthesized by the DNA Synthesis Laboratory, Department of Pathology, Yale University School of Medicine.

Detailed Description Text (74):

The PCR amplified gene fragment was ligated into the pCR.TM. vector using the TA cloning system (Invitrogen Co., San Diego, Calif.). Transformants were selected in LB medium containing 100 .mu.g/ml ampicillin and the plasmid DNA was isolated using the Wizard.TM. Minipreps DNA purification kit (Promega Co., Madison, Wis.). Insert DNA was released with the restriction enzymes EcoRI and XhoI (New England Biolabs, Beverly, Mass.), purified from an agarose gel electrophoresis and labeled with .sup.32 P using a random priming method (Megaprime Labeling Kit, Amersham Life Science, Buckinghamshire, England).

Detailed Description Text (75):

This DNA fragment was used as probe to screen a *L. donovani* promastigote cDNA library as described in Skeiky et al., Infect. Immun. 62:1643 (1994). An approximately 650 bp cDNA (Ldp23) was excised from the phagemid by in vivo excision using the Stratagene protocol. DNA sequencing was performed using the Sequenase version 2 system (DNA sequencing kit) in the presence or absence of 7-deaza-GTP (United States Biochemical, Cleveland, Ohio). The sequence is provided as SEQ ID NO:3, and shows complete homology with the original 300 bp PCR fragment. A 525 bp open reading frame containing an ATG codon that follows the last 4 bases of the spliced leader sequence and 3 stop codons adjacent to the poly A tail was identified. This frame also codes the carboxyl terminal sequence (KVFDE) (SEQ ID NO:13) of the purified MHC class II associated peptide. The sequence analysis of the deduced protein sequence revealed one potential glycosylation site (Asn-Cys-Ser) at positions 68-70.

Detailed Description Text (82):

To ascertain that the Ldp23 peptide is expressed in *Leishmania* organisms, a Northern blot analysis was performed using RNA prepared from different promastigote growth phases (logarithmic and stationary) and from the amastigote form of these

parasites.

Detailed Description Text (83):

The RNA was prepared from 2.times.10.sup.7 parasite cells using the Micro RNA isolation kit (Stratagene, La Jolla, Calif.) according to the company's recommended instructions. RNA was prepared from *L. donovani* promastigotes (logarithmic growth phase); from *L. major* promastigotes (logarithmic and stationary growth phases); from *L. amazonensis*, both promastigotes (logarithmic and stationary growth phases) and amastigotes purified from CBA/J infected mice; and from *L. pifanoi*, both promastigotes (logarithmic and stationary growth phases) and amastigotes (from axenic culture medium). *L. donovani* (1S strain), *L. amazonensis* (MHOM/BR/77/LTB0016), *L. major* (MHOM/IR/79/LRC-L251) and *L. pifanoi* (MHOM/VE/60/Ltrod) promastigotes were grown and maintained at 26.degree. C. in Schneider's medium containing 20% FCS and 50 .mu.g/ml gentamicin. The amastigote forms of *L. amazonensis* were obtained by differential centrifugation of a "pus-like" foot pad lesion of a CBA/J mouse infected for 6 months with this parasite. *L. pifanoi* amastigotes were obtained from axenic culture as previously reported by Pan et al., J. Euk. Microbiol. 40:213 (1993).

Detailed Description Text (85):

FIG. 4 shows that one single RNA band of 680 bp was observed for all growth phases and forms of all tested Leishmania. Within FIG. 4, the numbers 1, 2 and 3 refer to RNA obtained from promastigotes at the logarithmic growth phase, promastigotes at the stationary growth phase and amastigote forms, respectively, and the numbers on the left side indicate the molecular weights of the markers in base pairs. This result is consistent with the corresponding gene size (525 bp) and with the molecular weight of the expressed protein and points to the ubiquitous distribution and expression of this gene within the genus Leishmania.

Detailed Description Text (88):

Sera were prepared and the anti-Leishmania antibody response was measured by Western blot analysis and by FACScan. In both cases *L. donovani* promastigotes were used as antigen. Approximately 2.times.10.sup.6 *L. donovani* promastigotes were grown in Schneider's medium for 3 days (log phase), were washed with PBS, lysed with SDS-PAGE loading buffer and submitted to electrophoresis under reducing conditions using a 15% polyacrylamide gel. The proteins were transferred onto 0.45.mu. Immobilon-P transfer membrane (Millipore Co., Bedford, Mass.) using a wet-type electroblotter (Mini Trans-Blot Electrophoretic Transfer Cell, Bio Rad Life Science Division, Richmond, Calif.) for 2 hours at 50 V. The membranes were blocked overnight at room temperature with PBS containing 3% normal goat serum (NGS), 0.2% Tween-20 and 0.05% sodium azide, followed by 3 washes with PBS. The blots were then incubated for 3-4 hours at 4.degree. C. with a 1/200 dilution of pre-immune rabbit serum (lane A, FIG. 5) or with the same dilution of anti-fusion protein rabbit antiserum (lane B, FIG. 5). The sera was previously absorbed 2.times. with non-viable desiccated *Mycobacterium tuberculosis* H-37 RA (Difco Laboratories, Detroit, Mich.) and were diluted in PBS containing 1% NGS and 5% powdered non-fat bovine milk (Carnation, Nestle Food Company, Glendale, Calif.). The membranes were then washed with PBS, incubated for 1 hour at room temperature with goat anti-rabbit IgG antibody conjugated with alkaline phosphatase (Promega, Madison, Wis.), washed once with PBS and 2.times. with veronal buffer pH 9.4. The reaction was visualized using the substrate mixture 5-bromo-4-chloro-3-indoyl-phosphate and nitroblue tetrazolium (Kirkegaard & Perry Laboratories Inc., Gaithersburg, Md.) according to the manufacturer's instructions.

Detailed Description Text (89):

FIG. 5 shows that the rabbit anti-recombinant protein antiserum detects a single protein of 23 kDa (Ldp23) in the Leishmania crude extract antigen preparation. No bands were observed when an anti-GST antiserum was used (not shown). Moreover, the FACScan analysis (FIG. 6) shows that the antibody induced by the recombinant Ldp23 reacts with intact live *L. donovani* promastigotes, thus pointing to a cell surface expression of this molecule on these organisms. The dotted line in FIG. 6 shows the indirect immunofluorescence performed using pre-immune mouse serum and the solid line in FIG. 6 shows the result obtained with mouse anti-GST-Ldp23 antiserum. Both sera were diluted at 1/100. Parasites were washed with staining buffer and

h e b b g e e e f c e e ge

incubated with FITC conjugated goat anti-mouse immunoglobulin antibody. Fluorescence intensity was analyzed by FACScan.

Detailed Description Text (90):

F. Recognition of Recombinant Ldp23 by Leishmania-Specific Lymph Node T-cells

Detailed Description Text (91):

To test the responsiveness of T-cells to the Ldp23 protein, two sets of experiments were performed. In the first experiment, lymph node T-cells (10×10^5 /well) from BALB/c mice immunized with *L. donovani* promastigotes (as described above) were stimulated to proliferate with 2×10^5 Mitomycin C-treated normal mononuclear spleen cells (APC) and pulsed with the purified recombinant fusion protein. Proliferation of T-cells was measured at 72 hours of culture. Values are expressed in FIG. 7 as cpm and represent the mean of [3 H]TdR incorporation of triplicate cultures. Background cpm of cells (T cells+APC) cultured in the presence of medium alone was 1291. FIG. 7 shows that Leishmania specific T-cells proliferate well and in a dose response manner to recombinant Ldp23. No response was observed when purified GST was added instead of the recombinant fusion protein nor when lymph node T-cells from mice immunized with CFA alone were stimulated to proliferate in the presence of the Leishmanial fusion protein (not shown).

Detailed Description Text (92):

The recognition of the recombinant Ldp23 protein by Leishmania-specific T-cells was also tested using two murine models of leishmaniasis, the *L. major* highly susceptible BALB/c mice and the *L. amazonensis* susceptible CBA/J mice as described in Champs and McMahon-Pratt, Infect. Immun. 56:3272 (1988). These models were selected to investigate the cytokine pattern induced by Ldp23. In the mouse model of leishmaniasis, resistance is associated with Th 1 cytokines while susceptibility is linked to Th 2 responses.

Detailed Description Text (93):

Lymph node cells were obtained 3 weeks after the initiation of infection of BALB/c mice with *L. major* and the ability of these cells to recognize the recombinant Ldp23 was measured by proliferation and by the production of the cytokines IFN- γ and IL-4. 2×10^6 cells obtained from the draining popliteal lymph node of infected mice were cultured for 72 hours in the presence of recombinant Ldp23 or Leishmania lysate. The levels of IFN- γ and IL-4 in culture supernatants were measured by ELISA as previously described (Chatelain et al., J. Immunol. 148:1172 (1992), Curry et al., J. Immunol. Meth. 104:137 (1987), and Mossman and Fong, J. Immunol. Meth. 116:151 (1989)) using specific anti IFN- γ and IL-4 monoclonal antibodies (PharMingen, San Diego, Calif.).

Detailed Description Text (94):

Ldp23 did stimulate these cells to proliferate (not shown) and induced a typical Th 1 type of cytokine response as indicated by the production of high levels of IFN- γ (panel A of FIG. 8) and no IL-4 (panel B of FIG. 8). Stimulation of these cells with a Leishmania crude lysate yielded a mixed Th cytokine profile. Exactly the same pattern of cytokine production was obtained from the CBA/J mice infected with *L. amazonensis* (not shown). These results clearly indicate that Ldp23 is a powerful and selective activator of the Th 1 cytokines by mouse cells.

Detailed Description Text (97):

This Example illustrates the preparation of a Leishmania antigen Hsp83, having the sequence provided in SEQ ID NO:6.

Detailed Description Text (99):

Recombinant antigens produced by these clones were purified from 500 ml of isopropyl- β -D-thiogalactopyranoside (IPTG)-induced cultures as described in Skeiky et al., J. Exp. Med. 176:201-211 (1992). These antigens were then assayed for the ability to stimulate PBMC from Leishmania-infected individuals to proliferate and secrete cytokine. Peripheral blood was obtained from individuals living in an area (Corte de Pedra, Bahia, Brazil) where *L. braziliensis* is endemic and where epidemiological, clinical, and immunological studies have been performed for over a decade, and PBMC were isolated from whole blood by density

centrifugation through Ficoll (Winthrop Laboratories, New York, N.Y.). For in vitro proliferation assays, 2.times.10.sup.5 to 4.times.10.sup.5 cells per well were cultured in complete medium (RPMI 1640 supplemented with gentamicin, 2-mercaptoethanol, L-glutamine, and 10% screened pooled A+ human serum; Trimar, Hollywood, Calif.) in 96-well flat-bottom plates with or without 10 .mu.g of the indicated antigens per ml or 5 .mu.g of phytohemagglutinin per ml (Sigma Immunochemicals, St. Louis, Mo.) for 5 days. The cells were then pulsed with 1 .mu.Ci of [³H]thymidine for the final 18 h of culture. For determination of cytokine production 0.5 to 1 ml of PBMC was cultured at 1.times.10.sup.6 to 2.times.10.sup.6 cells per ml with or without the Leishmania antigens for 48 and 72 h.

Detailed Description Text (101):

A recombinant clone was identified in the above assays which, following sequence comparison of its predicted amino acid sequence with sequences of other proteins, was identified as a Leishmania braziliensis homolog of the eukaryotic 83 kD heat shock protein (Lbhsp83). The sequence of the clone is provided in SEQ ID NO:5 and the deduced protein sequence is provided in SEQ ID NO:6. On the basis of the homology, this clone, designated Lbhsp83a, appears to lack the first 47 residues of the full length 703 amino acid residues. Lbhsp83 has an overall homology of 94% (91% identity and 3% conservative substitution), 91% (84% identity and 7% conservative substitution) and 77% (61% identity and 16% conservative substitution) with L. amazonensis hsp83, T. cruzi hsp83 and human hsp89, respectively. A second clone (designated Lbhsp83b), which contained the 43 kD C-terminal portion of hsp83 (residues 331 to 703) was also isolated. FIG. 19 presents a comparison of the Lbhsp83 sequence with L. amazonensis hsp83(Lahsp83), T. cruzi hsp83 (Tchsp83) and human hsp89 (Huhsp89).

Detailed Description Text (102):

The results of proliferation assays using Lbhsp83a are shown in Table 1. Cells from all mucosal leishmaniasis (ML) patients proliferated strongly in response to Lbhsp83a, with stimulation indices (SIs) ranging from 19 to 558 (as compared to 20 to 1,634 for parasite lysate). Proliferation of PBMC from cutaneous leishmaniasis (CL) patients was variable and except for levels in two patients (IV and VII), levels were significantly lower than those of ML patients. By comparison, the proliferative responses of individuals with self-healing CL to Lbhsp83a were similar to those of individuals with ML. However, the responses of all six self-healing individuals to Lbhsp83 were consistently higher than those to Lbhsp83b. This suggests that PBMC from self-healing CL patients preferentially recognize one or more T-cell epitopes located within the amino portion of Lbhsp83.

Detailed Description Text (104):

In PBMC of three ML patients, stimulation with lysate resulted in increased expression of mRNA for IFN-.gamma., IL-2, and IL-4 but not IL-10. By comparison, both Lbhsp83 polypeptides elicited the production of mRNA for IFN-.gamma. and IL-2 from all ML patient PBMC tested. In contrast, profiles of mRNA for IL-10 and IL-4 differed for the two hsp83 polypeptides. Lbhsp83a stimulated the production of IL-10 but not IL-4 mRNA (patients I, II, III, and IV), while Lbhsp83b stimulated the production of IL-4 but not IL-10 mRNA in all six patients.

Detailed Description Text (105):

All CL patients tested responded to both Lbhsp83 polypeptides as well as to the parasite lysate by upregulating the synthesis of mRNAs for IL-2 and IFN-.gamma., and in two of four patients (I and IV), the level of IL-4 mRNA also increased, indicating stimulation of both Th1 and Th2 cytokines. Interestingly and as in the case of ML patient uncultured PBMC which did not have detectable levels of IL-10 mRNA, Lbhsp83a and not Lbhsp83b stimulated PBMC from one CL patient (IV) to synthesize IL-10 mRNA. However, in the other three patients (I, II, and III) with resting levels of IL-10 mRNA, both rLbhsp83 polypeptides as well as the parasite lysate downregulated the expression of IL-10 mRNA.

Detailed Description Text (106):

PBMC supernatants were also assayed for the presence of secreted IFN-.gamma., TNF-.alpha., IL-4, and IL-10. Cells from all ML and self-healing CL patients (seven and

six patients, respectively) and from four of seven CL patients were analyzed for secreted IFN- γ . following stimulation with both rLbhsp83 polypeptides, parasite lysate and Lbhsp70, an *L. braziliensis* protein homologous to the eukaryotic 70 kD heat shock protein (FIG. 10A). In general, rLbhsp83a stimulated patient PBMC to secrete higher levels of IFN- γ . than did rLbhsp83b (0.2 to 36 and 0.13 to 28 ng/ml, respectively). The presence of secreted IFN- γ . correlated well with the corresponding mRNA detected by PCR.

Detailed Description Text (107):

PBMC from four of five ML patients (I, II, V, and VII) had supernatant TNF- α . levels (0.8 to 2.2 ng/ml) higher than those detected in cultures of PBMC from uninfected controls following stimulation with parasite lysate (FIG. 10B). Similarly, the same PBMC were stimulated by rLbhsp83 to produce levels of TNF- α . in supernatant ranging from 0.61 to 2.9 ng/ml. Compared with those of uninfected controls, PBMC from three (I, V, and VI), five (I, II, IV, V, and VI), and two (II and V) of six individuals analyzed produced higher levels of TNF- α . in response to parasite lysate, rLbhsp83a, and rLbhsp83b, respectively. The levels of TNF- α . produced by PBMC from CL patients in response to parasite lysate were comparable to those produced by uninfected controls. However, rLbhsp83 stimulated TNF- α . production in the PBMC of two of these patients. rLbhsp83a stimulated higher levels of TNF- α . production than did rLbhsp83b. In the absence of antigen stimulation, only PBMC from ML patients (five of six) produced detectable levels of supernatant TNF- α . (60 to 190 pg/ml).

Detailed Description Text (108):

In agreement with the IL-10 mRNA, IL-10 was detected by ELISA in the antigen-stimulated PMBC culture supernatants from ML and CL patients. The levels (49 to 190 pg) were significantly higher (up to 10-fold) following stimulation with rLbhsp83a compared with those after parallel stimulation of the same cells with rLbhsp83b (FIG. 11). Parasite lysate also stimulated PMBC from some of the patients to produce IL-10. Although rLbhsp83 stimulated PMBC from uninfected individuals to produce IL-10, with one exception, the levels were lower than those observed with patient PMBC. IL-4 was not detected in any of the supernatants analyzed. Therefore, the level of any secreted IL-4 is below the detection limit of the ELISA employed (50 pg/ml). Taken together, the results demonstrate that a predominant Th1-type cytokine profile is associated with PMBC from *L. braziliensis*-infected individuals following stimulation with rLbhsp83 polypeptides.

Detailed Description Text (109):

To determine the correlation between the observed T-cell responses and antibody production to Lbhsp83, we compared the antibody (immunoglobulin G) reactivities to Lbhsp83 in sera from the three patient groups (FIG. 12). The ELISA reactivities of ML patient sera with rLbhsp83a were comparable to those observed with parasite lysate, and in general, there was a direct correlation between ML patient anti-Lbhsp83 antibody titer and T-cell proliferation. Of 23 serum samples from ML patients analyzed, 22 were positive (.about.96%) with absorbance values of 0.20 to >3.0. Eleven of the ML patient serum samples had optical density values that were >1. In general, CL patients had significantly lower anti-Lbhsp83 antibody titers ({character pullout}=0.74; standard error of the mean [SEM]=0.1) compared to those of ML patients. Therefore, ML and CL patient anti-rhsp83 antibody titers correlated with their respective T-cell proliferative responses. Anti-rLbhsp83 antibody titers were significantly higher in patients with ML ({character pullout}=1.5; SEM=0.2) than in self-healing CL patients ({character pullout}=0.35; SEM=0.056), although their T-cell proliferative responses were similar. In fact, anti-Lbhsp83 antibody titers in serum from self-healing CL patients were comparable to those from uninfected controls ({character pullout}=0.24; SEM=0.028). By using 2 standard deviations greater than the mean absorbance value of uninfected control (0.484) as a criterion for positive reactivity to Lbhsp83, eight of nine of the self-healing patient serum samples tested were negative.

Detailed Description Text (112):

This Example illustrates the preparation of clones encoding portions of the Leishmania antigen Lt-210, and which has the sequence provided in SEQ ID NO:8.

Detailed Description Text (119):

Hybridization analysis confirmed that rLt-2 and rLt-1 contain overlapping sequences. Genomic DNAs of various Leishmania species were restricted with a variety of enzymes, separated by agarose gel electrophoresis, and blotted on Nytran membrane filter (Schleicher & Schuell, Keene, N.H.). Inserts from rLt-1 and rLt-2 were labeled with 32 P-CTP by reverse transcriptase from random oligonucleotide primers and used as probes after separation from unincorporated nucleotides on a Sephadex G-50 column. Hybridizations using the rLt-1 or the rLt-2 probe are performed in 0.2M NaH.sub.2 PO.sub.4 /3.6 M NaCl at 65.degree. C., whereas hybridization using the rLt-1r probe is performed in 0.2 M NaH.sub.2 PO.sub.4 /3.6 M NaCl/0.2 M EDTA at 60.degree. C. overnight. Filters are washed in 0.075 M NaCl/0.0075 M sodium citrate pH 7.0 (0.15 M NaCl/0.0150 M sodium citrate for the Lt-1r probe), plus 0.5% SDS at the same temperature as hybridization.

Detailed Description Text (120):

Genomic DNA from a number of Leishmania species including *L. tropica* were analyzed by Southern blots as described above using the Lt-1, Lt-2, and Lt-1r inserts separately as probes. Collectively, various digests of *L. tropica* DNA indicate that this gene has a low copy number. A similar, overlapping pattern was observed using either the Lt-1 or Lt-2 insert as a probe, consistent with the premise that these two clones contain sequences near or overlapping one another. In addition, sequences hybridizing with these clones are present in other Leishmania species.

Detailed Description Text (128):

A recombinant clone was identified in the above assays which, following sequence comparison of its predicted amino acid sequence with sequences of other proteins, was identified as a Leishmania braziliensis homolog of the eukaryotic initiation factor 4A (eIF4A). The isolated clone (pLeIF.1) lacked the first 48 amino acid residues (144 nucleotides) of the full length protein sequence. The pLeIF.1 insert was subsequently used to isolate the full length genomic sequence.

Detailed Description Text (132):

This Example illustrates the preparation of soluble Leishmania antigens from an *L. major* culture supernatant. *L. major* promastigotes were grown to late log phase in complex medium with serum until they reached a density of 2-3.times.10.sup.7 viable organisms per mL of medium. The organisms were thoroughly washed to remove medium components and resuspended at 2-3.times.10.sup.7 viable organisms per mL of defined serum-free medium consisting of equal parts RPMI 1640 and medium 199, both from Gibco BRL, Gaithersburg, Md. After 8-12 hours, the supernatant was removed, concentrated 10 fold and dialyzed against phosphate-buffered saline for 24 hours. Protein concentration was then determined and the presence of at least eight different antigens confirmed by SDS-PAGE. This mixture is referred to herein as "soluble Leishmania antigens."

Detailed Description Text (134):

Comparison of Interleukin-4 and Interferon-.gamma. Production Stimulated by Leishmania Antigens

Detailed Description Text (135):

This Example illustrates the immunogenic properties of the antigens prepared according to Examples 1, 2, 5 and 6, as determined by their ability to stimulate IL-4 and IFN-.gamma. in lymph node cultures from infected mice and in human PBMC preparations. Lymph node cultures for use in these studies were prepared from *L. major*-infected BALB/c mice 10 days after infection, as described in Example 2. PBMC were prepared using peripheral blood obtained from individuals with cured *L. donovani* infections who were immunologically responsive to Leishmania. Diagnosis of the patients was made by clinical findings associated with at least one of the following: isolation of parasite from lesions, a positive skin test with Leishmania lysate or a positive serological test. Uninfected individuals were identified based on a lack of clinical signs or symptoms, a lack of history of exposure or travel to endemic areas, and the absence of a serological or cellular response to Leishmania antigens. Peripheral blood was collected and PBMC isolated by density centrifugation through Ficoll.TM. (Winthrop Laboratories, New York).

Detailed Description Text (137):

FIGS. 13A and 13B, illustrate the mean level of secreted IL-4 and IFN- γ , respectively, 72 hours after addition of 10 $\mu\text{g/mL}$ of each of the following antigens to a lymph node culture prepared as described above: soluble Leishmania antigen (i.e., an extract prepared from ruptured promastigotes which contains membrane and internal antigens (SLA)), Ldp23, LbeIF4A (LeIF), Lbhsp83, M15 and LmeIF (the L. major homolog of LbeIF4A). The levels of secreted IL-4 and IFN- γ in medium alone (i.e., unstimulated) are also shown. While SLA elicits a predominantly Th2 response from lymph node cells of Leishmania-infected mice, Ldp23, LbeIF4A, Lbhsp83 and M15 elicited relatively little IL-4 and large amounts of IFN- γ , consistent with a Th1 response profile.

Detailed Description Text (138):

FIG. 14 shows the level of secreted IFN- γ in culture filtrate from infected and uninfected human PBMC preparations 72 hours after addition of 10 $\mu\text{g/mL}$ L. major lysate, M15 or L-Rack, an immunodominant leishmanial antigen in murine leishmaniasis. Similarly, FIG. 15 illustrates the level of secreted IFN- γ in culture filtrate from infected and uninfected human PBMC preparations 72 hours after addition of 10 $\mu\text{g/mL}$ L. major lysate, soluble Leishmania antigens (prepared as described in Example 6) or L-Rack. These results indicate that M15 and soluble Leishmania antigens, but not L-Rack, are potent stimulators of IFN- γ production in patient PBMC, but not in PBMC obtained from uninfected individuals. Thus, M15 and soluble Leishmania antigens elicit a dominant Th1 cytokine profile in both mice and humans infected with Leishmania.

Detailed Description Text (140):

Comparison of Proliferation Stimulated by Leishmania Antigens

Detailed Description Text (143):

FIG. 16 illustrates the proliferation observed after addition of 10 $\mu\text{g/mL}$ or 20 $\mu\text{g/mL}$ of each of the following antigens to a lymph node culture prepared as described in Example 7: SLA, Ldp23, LbeIF4A, Lbhsp83, and M15. The level of proliferation without the addition of antigen is also shown. Data are represented as mean cpm. These results demonstrate that a variety of leishmanial antigens are capable of stimulatory lymph node cell proliferation from Leishmania-infected mice.

Detailed Description Text (144):

FIGS. 17 and 18 illustrate the proliferation observed in human PBMC preparations from Leishmania-immune and uninfected individuals following the addition of 10 $\mu\text{g/mL}$ M15 and soluble Leishmania antigens, respectively. These values are compared to the proliferation observed following the addition of culture medium, L. major lysate or L-Rack. The results show that M15 and soluble Leishmania antigens stimulate proliferation in Leishmania-immune PBMC, but not in PBMC obtained from uninfected individuals, demonstrating that M15 and soluble antigens (but not L-Rack) are recognized by PBMC from individuals immune to Leishmania due to a previous infection.

Detailed Description Text (147):

This Example illustrates the preparation of two soluble Leishmania antigens, Lmspla and Lmsp9a.

Detailed Description Text (150):

Anti E. coli antibody reactivities were removed from the rabbit sera by pre-adsorbing on nitrocellulose filters containing lysed E. coli. Adsorbed sera were evaluated by Western blot analysis using 10 μg Leishmania promastigote lysate (lane 1) and 1 μg soluble L. major antigen mixture (lane 2). As shown in FIG. 20, the rabbit sera was found to be reactive with seven dominant antigens of the soluble L. major antigen mixture with molecular weights ranging from 18 to >200 kDa. A four times longer exposure of the same blot revealed three additional immunoreactive species with molecular weights less than 18 kDa. The same sera reacted with approximately 10 antigens of the promastigote lysate, but with a pattern significantly different from that observed with the soluble L. major antigens (FIG. 20). This is suggestive of potential post-translational modification

of the same antigen before (intracellular localization) and after secretion/shedding. Such modifications may include cleavage of a leader sequence and/or the addition of carbohydrate molecules to the secreted/shed antigens.

Detailed Description Text (151):

The rabbit sera described above was subsequently used to screen an L. major cDNA expression library prepared from L. major promastigote RNA using the unidirectional Lambda ZAP (uni-ZAP) kit (Stratagene) according to the manufacturer's protocol. A total of 70,000 pfu of the amplified cDNA library was screened with the rabbit sera at a 1:250 dilution. Nineteen positive clones were confirmed in the tertiary screening. The phagemid were excised and DNA from each of the 19 clones was sequenced using a Perkin Elmer/Applied Biosystems Division automated sequencer Model 373A. All 19 clones were found to represent two distinct sequences, referred to as Lmspla and Lmsp9a. The determined cDNA sequences for Lmspla and Lmsp9a are provided in SEQ ID NO: 19 and 21, respectively, with the corresponding amino acid sequences being provided in SEQ ID NO: 20 and 22, respectively.

Detailed Description Text (155):

Southern blot analysis of genomic DNA from L. major (Friedlander strain) digested with a panel of restriction enzymes (lanes 1 to 7) and six other Leishmania species of different geographic locations digested with PstI (lanes 8 to 13) using the full-length cDNA insert of Lmspla, demonstrated that Lmspla is present in all the species characterized with a high degree of conservation (FIG. 22). This suggests evolutionary significance for the maintenance of Lmspla and the existence of homologous species among all the Leishmania species.

Detailed Description Text (156):

The remaining two cDNA clones isolated from the soluble L. major antigen mixture represent identical sequences (referred to as Lmsp9a; SEQ ID NO: 21), suggesting that the two copies resulted from amplification of the primary library. Sequencing of the Lmsp9a cDNA revealed that the clone does not contain the full length 5' sequence since it is lacking both the spliced leader and 5' untranslated sequences. The 3' end of the cDNA contains a poly A stretch, as would be expected for a Leishmania mRNA. Of the predicted translated sequence (SEQ ID NO: 22), 34 of the 201 amino acids (17%) represent cysteine residues. Comparison of the predicted protein sequence with those of known proteins as described above, revealed some homology with other cysteine rich proteins such as the major surface trophozoite antigen of Giardia lamblia and furin proteases.

Detailed Description Text (159):

This Example illustrates the preparation and characterization of the Leishmania antigen MAPS-1A (SEQ ID NO: 24).

Detailed Description Text (161):

One of these clones, referred to herein as MAPS-1A, was found to be full-length. Comparison of the cDNA and deduced amino acid sequences for MAPS-1A (SEQ ID Nos: 23 and 24, respectively) with known sequences in the gene bank using the DNA STAR system revealed no significant homologies to known Leishmania sequences, although some sequence similarity was found to a group of proteins, known as thiol-specific antioxidants, found in other organisms.

Detailed Description Text (162):

Recombinant MAPS-1A protein having an amino-terminal HIS-Tag was prepared using a high level E. coli expression system and recombinant protein was purified by affinity chromatography as described in Example 1. Southern blot analysis of genomic DNA from L. major digested with a panel of restriction enzymes, seven other Leishmania species digested with PstI, and two other infectious-disease pathogens (T. cruzi and T. brucei), using the full length insert of MAPS-1A, demonstrated that MAPS-1A is present in all eight Leishmania species tested (FIG. 23). Northern blot analysis of L. major promastigote and amastigote RNAs indicated that MAPS-1A is constitutively expressed.

Detailed Description Text (164):

The ability of recombinant MAPS-1A to stimulate cell proliferation was investigated

as follows. PBMC from 3 *L. braziliensis*-infected patients having active mucosal leishmaniasis, from 4 patients post kala-azar infection (previously infected with *L. chagasi* and/or *L. donovani*) and from 3 uninfected-individuals were prepared as described above in Example 7. The ability of MAPS-1A to stimulate proliferation of these PBMC was determined as described in Example 8 above. As shown in FIG. 24, significant levels of MAPS-1A specific PBMC proliferation were seen in 2 of the 7 *Leishmania* patients.

Detailed Description Text (167):

Immunoreactivity of Soluble *Leishmania* Antigens with Sera from *Leishmania*-Infected Patents

Detailed Description Text (171):

As shown in FIG. 26, approximately 50% of the samples from human leishmaniasis patients showed reactivities with recombinant MAPS-1A substantially above background. FIG. 27 shows the reactivity of MAPS-1A with increasing dilutions of sera from BALB/c mice previously administered either (i) saline solution; (ii) the adjuvant B. pertussis; (iii) soluble *Leishmania* antigens plus B. pertussis; (iv) live *L. major* promastigotes; or (v) soluble *Leishmania* antigens plus B. pertussis followed by live *L. major* promastigotes (as described below in Example 12). Considerably higher absorbances were seen with sera from mice infected with live *L. major* promastigotes and with mice infected with live *L. major* promastigotes following immunization with soluble *Leishmania* antigens plus B. pertussis, than with sera from the other three groups of mice, indicating that anti-MAPS-1A antibody titers increase following *Leishmania* infection.

Detailed Description Text (173):

Use of *Leishmania* Antigens for Vaccination Against *Leishmania* Infection

Detailed Description Text (174):

This example illustrates the effectiveness of *Leishmania* antigens in conferring protection against disease in the experimental murine leishmaniasis model system. For a discussion of the murine leishmaniasis model system see, for example, Reiner et al. Annu. Rev. Immunol., 13:151-77, 1995.

Detailed Description Text (175):

The effectiveness of (i) crude soluble *Leishmania* antigens, (ii) MAPS-1A, and (iii) a mixture of Ldp23, LbeIF4A and M15, as vaccines against *Leishmania* infection was determined as follows. BALB/c mice (5 per group) were immunized intra-peritoneally three times at biweekly intervals with either (i) 30 .mu.g crude soluble *Leishmania* antigens, (ii) 20 .mu.g MAPS-1A or (iii) a mixture containing 10 .mu.g each of LeIF, Ldp23 and M15, together with 100 .mu.g of the adjuvant C. parvum. Two control groups were immunized with either saline or C. parvum alone. Two weeks after the last immunization, the mice were challenged with 2.times.15.sup.5 late-log phase promastigotes of *L. major*. Infection was monitored weekly by measurement of footpad swelling. The amount of footpad swelling seen in mice immunized with either crude soluble *Leishmania* antigens, a mixture of Ldp23, LbeIF4A and M15 (FIG. 28), or MAPS-1A (FIG. 29) was significantly less than that seen in mice immunized with C. parvum alone. These results demonstrate that the *Leishmania* antigens of the present invention are effective in conferring protection against *Leishmania* infection.

Detailed Description Text (178):

This example illustrates the isolation of seven soluble *Leishmania* antigen genes from an *L. major* genomic DNA library.

Detailed Description Text (179):

An *L. major* genomic DNA expression library was prepared from *L. major* promastigotes using the unidirectional Lambda ZAP (uni-ZAP) kit (Stratagene) according to the manufacturer's protocol. This library was screened with a high titer rabbit sera raised against *L. major* soluble antigens, as described above in Example 9. Seven positive clones were identified. The phagemid were excised and DNA from each of the seven clones was sequenced using a Perkin Elmer/Applied Biosystems Division automated sequencer Model 373A. The DNA sequences for these antigens, referred to as LmgSP1, LmgSP3, LmgSP5, LmgSP8, LmgSP9, LmgSP13, LmgSP19, are provided in SEQ ID

NO:29-35, respectively, with the corresponding amino acid sequences being provided in SEQ ID NO: 36-42, respectively. LmgSP13 was found to contain a 39 amino acid repeat sequence shown in SEQ ID NO:43.

Detailed Description Text (182):

The reactivity of recombinant LmgSP9 with sera from patients with visceral leishmaniasis, (from both Sudan and Brazil) and from normal donors was evaluated by ELISA as described above. The absorbance values were compared with those obtained using the known Leishmania antigen K39 described above, with L. chagasi lysate being employed as a positive control. Representative results of these assays are provided below in Table 2, wherein all the patients from Brazil and those from the Sudan designated as "VL" were inflicted with visceral leishmaniasis. The results demonstrated that LmgSP9 specifically detects antibody in most individuals with visceral leishmaniasis, regardless of geographical location. In several cases, the absorbance values of the antibody reactivity to LmgSP9 were comparable to that observed with K39. In addition, LmgSP9 detected several cases of leishmaniasis that were not detected using K39. These results indicate that LmgSP9 can be used to complement the reactivity of K39.

Detailed Description Text (188):

This example illustrates the preparation of five soluble Leishmania antigen genes from an L. chagasi genomic DNA library.

Detailed Description Text (189):

An L. chagasi genomic DNA expression library was prepared from L. chagasi promastigotes using the unidirectional Lambda ZAP (uni-ZAP) kit (Stratagene) according to the manufacturer's protocol. This library was screened with a high titer rabbit sera raised against L. major soluble antigens, as described above in Example 9. Five positive clones were identified. The phagemid were excised and DNA from each of the Five clones was sequenced using a Perkin Elmer/Applied Biosystems Division automated sequencer Model 373A. The DNA sequences for these antigens, referred to as LcgSP1, LcgSP3, LcgSP4, LcgSP8, and LcgSP10 are provided in SEQ ID NO:44-48, respectively, with the corresponding amino acid sequences being provided in SEQ ID NO:49-53, respectively.

Detailed Description Text (195):

Leishmania-specific CD4+ T cell lines were derived from the PBMC of an individual who tested positive in a leishmania skin test but had no clinical history of disease. These T cell lines were used to screen a L. major amastigote cDNA expression library prepared as described in Example 1. Immunoreactive clones were isolated and sequenced as described above. The determined cDNA sequences for the 8 isolated clones referred to as 1G6-34, 1E6-44, 4A5-63, 1B11-39, 2A10-37, 4G2-83, 4H6-41, 8G3-100 are provided in SEQ ID NO: 72-79. respectively, with the corresponding predicted amino acid sequences being provided in SEQ ID NO: 80-87, respectively. The cDNA sequences provided for 1E6-44, 2A10-37, 4G2-83, 4H6-41 and 8G3-100 are believed to represent partial clones. All of these clones were shown to stimulate T cell proliferation.

Detailed Description Text (201):

Use of Leishmania Antigens Plus Adjuvant for Vaccination Against Leishmania Infection

Detailed Description Text (202):

This example illustrates the effectiveness of recombinant Leishmania antigens, M15 and MAPS, plus an adjuvant, IL-12, in conferring protection against disease in the experimental murine leishmaniasis model system. For discussion of the murine leishmaniasis model system see, for example, Reiner et al., *Annu. Rev. Immunol.*, 13:151-77, 1995. The effectiveness of M15 and MAPS in combination with IL-12, as vaccine against Leishmania infection was determined as follows: BALB/c mice (5 per group) were immunized subcutaneously in the left footpad, twice (3 weeks apart) with the 10 .mu.g of the individual antigens mixed with 1 .mu.g of IL-12. As controls, three separate groups of mice were immunized with soluble leishmania lysate antigens (SLA) plus IL-12, with IL-12 alone or with PBS. Three weeks after the last immunization the mice were infected in the right footpad with

2.times.10.sup.5 promastigote forms of *L. major* (stationary phase). Footpad swelling was then measured weekly. Results are expressed in FIG. 31 and clearly indicate that the mice immunized with either M15 or MAPS and IL-12 were greatly protected against the infection; whereas mice immunized with IL-12 alone did not show protection from infection. The protection induced by these antigens was as efficient or better than that induced by SLA+IL-12, a regimen known to induce good protection against leishmaniasis in this animal model (Afonso, L. C. C., T. M. Scharon, L. Q. Vieira, M. Wysocka, G. Trinchieri, and P. Scott. 1994. The adjuvant effect of interleukin-12 in a vaccine against *Leishmania major*. *Science* 263:235-237). The same pattern of protection described above, was obtained i.e., M15, MAPS, and SLA, induced protection against *L. major* infection when *C. parvum* instead of IL-12 was used as adjuvant (Example 12). These results demonstrate that both M15 and MAPS recombinant antigens induce excellent protection against *L. major* infection in the BALB/c model of human leishmaniasis. In addition, both antigens induced protection when tested in two different adjuvant formulations, (e.g., IL-12 and *C. parvum*.) This finding is of high significance because it demonstrates that immunity to leishmaniasis can be induced by the specific antigens delivered in adjuvants that are suitable for human use.

Detailed Description Text (204):

Use of Leishmania DNA for Vaccination Against Leishmania Infection

Detailed Description Text (205):

This example illustrates the effectiveness of Leishmania DNA in conferring protection against disease in the experimental murine leishmaniasis model system. For discussion of the murine leishmaniasis model system see, for example, Reiner et al., *Annu. Rev. Immunol.*, 13:151-77, 1995. The protection properties of the recombinant antigens was tested by immunizing mice with naked DNA containing the corresponding M15 and MAPS genes. The DNA construct used was the pcDNA3.1 vector (Invitrogen) containing a CMV promoter. BALB/c mice (5 per group) were injected in the left footpad three times (3 weeks apart) with 100 .mu.g of the indicated naked DNA preparations. Mice were bled before and after the immunizations to monitor the development of specific immune response. The antibody response was evaluated by ELISA. Specific anti-M15 and anti-MAPS IgG2a antibodies were detected after the second immunization in the sera of the mice immunized with the respective naked DNA. The presence of specific antibodies indicates that the DNA immunization resulted in the production of specific protein antigen. Three weeks after the last immunization, the mice were then challenged in the right footpad with 2.times.10.sup.5 promastigote forms of *L. major* (stationary phase). Footpad swelling was then measured weekly thereafter. Results are expressed in FIG. 32 and clearly indicated that, again, mice immunized with naked DNA containing either the M15 or MAPS genes were greatly protected against the infection with *L. major*. These results demonstrate that both M15 and MAPS genes induce excellent protection against *L. major* infection in the BALB/c model of human leishmaniasis.

Detailed Description Text (207):

Preparation and Characterization of Leishmania Fusion Proteins

Detailed Description Text (208):

Fusion proteins comprising the Leishmania antigens MAPS-1A (SEQ ID NO: 24), M15 (SEQ ID NO: 2), Lbhsp83 (SEQ ID NO: 6) and LbeIF4A (SEQ ID NO: 10) were prepared as follows.

Detailed Description Text (214):

Use of Leishmania Fusion Proteins Plus Adjuvant for Vaccination Against Leishmania Infection

Detailed Description Text (215):

The ability of the Leishmania fusion proteins MAPS1A-M15 (referred to as the diFusion) and MAPS1A-M15-LbeIF4A (referred to as the triFusion), plus adjuvant, to confer protection against disease in the experimental murine leishmaniasis model system was examined as follows.

Detailed Description Paragraph Table (1):

h e b b g e e f c e e ge

TABLE 1 In vitro Proliferation of PMBC from *L. braziliensis*-infected Individuals in Response to Lbhsp83 Mean [10^{sup.3} H] thymidine Group and incorporation [10^{sup.3} cpm (SD)], SI with: Patient Lysate Lbhsp83a Lbhsp83b ML I 41.3, (1.3), 294 32.5, (6.6), 221 46.7, (1.4), 318 II 44.2, (0.5), 104 20, (3.7), 47 36.7, (0.76), 86 III 27.4, (1.5), 150 8.1, (1.7), 44 9.9, (0.32), 54 IV 52.7, (3.3), 138 54.1, (6.2), 142 32.0, (1.3), 84 V 140.6, (7.6), 308 151.8, (57), 333 150.4, (7.9), 331 VI 15.8, (1.8), 20 21.3, (4.4), 28 14.4, (1.3), 19 VII 300.1, (9.4), 1634 102.1, (7.6), 558 41.7, (4.9), 228 CL I 0.26, (0.0), 1.5 0.57, (0.3), 3.3 0.43, (0.17), 3.3 II 55.63, (8.6), 218 0.42, (0.0), 1.6 0.8, (0.14), 3.2 III 0.39, (0.5), 4.0 3.4, (0.5), 9 2.6, (0.9), 6.6 IV 19.14, (1.3), 87 7.17, (0.6), 32 5.9, (0.9), 27 V 0.32, (0.2), 3.0 1.47, (0.5), 14 0.3, (0.1), 3.0 VI 0.77, (0.1), 4.7 1.44, (0.2), 9 1.3, (0.6), 8.0 VII 4.01, (1.0), 2.0 60.3, (8.5), 15 66.7, (3.9), 16.6 Self-healing CL I 19.7, (4.4), 94 61.3, (4.6), 293 5.0, (2.0), 24 II 0.6, (0.1), 6.5 7.0, (2.0), 79 1.2, (0.8), 13 III 59.6, (7.1), 519 49.4, (3.1), 429 21.4, (3.7), 186 IV 0.2, (0.1), 1.6 13.1, (1.7), 108 0.6, (0.1), 5 V 27.1, (2.0), 225 6.3, (2.6), 52 3.0, (1.5), 25 VI 130.3, (14), 340 28.2, (2.9), 74 7.7, (3.8), 20 Control (uninfected) I 0.19, (0.0), 1.4 0.18, (0.0), 1.3 0.40, (0.16), 2.8 II 0.31, (0.1), 1.7 0.19, (0.0), 1.0 0.27, (0.0), 1.5 III 0.44, (0.2), 4.1 0.48, (0.1), 5.0 0.51, (0.2), 5.2 IV 0.4, (0.1), 3.2 0.52, (0.2), 5.1 0.50, (0.1), 5.0

Detailed Description Paragraph Table (2):

TABLE 2 REACTIVITY OF LMGSP9 WITH SERA FROM LEISHMANIA PATIENTS Patient No. L. chagasi lysate K39 LmgSP9 Sudanese samples: B19 1.067 0.306 0.554 B25 1.884 3.435 0.974 B43 1.19 3.225 0.86 B47 2.405 2.892 0.375 B50 0.834 0.748 0.432 B58 0.921 0.235 0.92 B63 1.291 0.303 0.764 B70 0.317 0.089 3.056 VL4 1.384 3.035 2.965 VL11 0.382 0.144 0.142 VL12 0.277 0.068 0.098 VL13 0.284 0.12 0.194 Brazilian samples: 105 3.508 3.53 0.374 106 2.979 3.373 2.292 107 2.535 3.444 0.46 109 1.661 3.415 3.319 111 3.595 3.537 0.781 112 2.052 3.469 0.63 113 3.352 3.429 0.963 114 2.316 3.437 1.058 115 2.073 3.502 1.186 116 3.331 3.461 0.96 Normal Donors: 129 0.157 0.104 0.08 130 0.195 0.076 0.095 131 0.254 0.134 0.086 132 0.102 0.035 0.043

Detailed Description Paragraph Table (3):

SEQUENCE LISTING <100> GENERAL INFORMATION: <160> NUMBER OF SEQ ID NOS: 101 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 1 <211> LENGTH: 3134 <212> TYPE: DNA <213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222> LOCATION: (1)...(3134) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 1 caagtgtcga aggacagtgt tcncctgtgt agatcgccgg ctgtgctgt gaaggcgggtg 60 ccatcggana aacaacaccg gtgganccgc aggaaccat ctttctccgc aggtctcttt 120 ttgttgtcga ttgagagtgc nccaaaccct gctggtgcc ttctcacata tcatgttttt 180 cgttgtgccc tcgctttgcc tttcctctcc tttccctctc ttccgtggtg ccgtgtatac 240 ttctggcacc cgctacgtca cttcgctggt ttgaacagaa ccaactgtgaa caccacggg 300 cgatcgaca catacacatc cctcactcac acacacagct acatctatcc tacataaagc 360 tgaaaaaaa gtctacgaac aattttgttt ttacagtgcg ttgccgcaca tttctccgta 420 atggacgcaa ctgagctgaa gaacaagggt aacgaagagt tctccgcccg ccgctatgtg 480 gaggcgggtga actacttctc aaaggcgatc cagttggatg agcagaacag tgtcctctac 540 agcaaccgct ccgcctgttt tgcagccatg cagaaataca aggacgcgct ggacgacgcc 600 gacaagtgca tctcgatcaa gccgaattgg gccaaagggt acgtgcggcg aggagcagct 660 ctccatggca tgcgccgcta cgacgatgcc attgccgcgt atgaaaagg gctcaagggt 720 gacccttcca acagcggtg cgcgaggggc gtgaaggacg tgcaggtagc caaggccgc 780 gaagcacgtg accccatcgc tcgctcttc accccggagg cgttccgcaa gatccaagag 840 aatcccaagc tgtctctact tatgtgcag ccgactacg tgaagatggt agacaccgtc 900 atccgcgacc cttcgcaggg ccggtgttac atggaagacc agcgctttgc cctgacgctc 960 atgtacctga gcggaatgaa gattcccaac gatggtgatg gcgaggagga ggaacgtccg 1020 tctgcgaagg cggcagagac agcgaagcca aaagaggaga agcctctcac cgacaacgag 1080 aaggaggccc tggcgctcaa ggaggagggc aacaagctgt acctctcgaa gaagtttgag 1140 gaggcgctga ccaagtacca agaggcgagc gtgaaagacc ccaacaacac ttatatacatt 1200 ctgaacgtgt cggccgtgta cttcgagcag ggtgactacg acaagtgcac cgccgagtgc 1260 gagcacggta tcgagcacgg tcgcgagAAC cactgcgact acacaatcat tgcgaagctc 1320 atgaccggga acgccttggt cctccagagg cagagggaagt acgaggctgc tatcgacctt 1380 tacaagcgcg cccttgctga gtggcgtaac cctgacaccc tcaagaagct gacggagtgc 1440 gagaaggagc accaaaaggc ggtggaggaa gcctacatcg atcctgagat cgcgaagcag 1500 aagaagacg aaggtaacca gtacttcaag gaggataagt tccccgaggc cgtggcagcg 1560 tacacggagg ccatcaagcg caaccctgcc gagcacacct cctacagcaa tcgcgcggcc 1620 gcgtacatca agcttgagc cttcaacgac gccctcaagg acgcggagaa gtgcattgag 1680 ctgaagcccg actttgttaa gggctacgcg cgcaagggtc atgcttactt ttggaccaag 1740 cagtacaacc gcgcgctgca ggcgtagcat gagggcctca aggtggaccc gagcaatgcg 1800 gactgcaagg atgggcggta tcgcacaatc atgaagattc aggagatggc atctggccaa 1860 tccgcggatg gcgacgaggc ggcgcggcgg gccatggacg atcctgaaat cgcggcaatc 1920

atgcaagata gctacatgca actagtgttg aaggagatgc agaacgatcc cacgcgcatt 1980 caggagtaca
tgaaggactc cggtatctca tcgaagatca acaagctgat ttcagctggc 2040 atcattcgtt ttggtcagta
gacttctacg ctgcctcatc ttttccgtgt ctttgcgtcg 2100 gcgggtatcg taaagcaca taaagcagcg
attcacatgc acgagtaaag tgctgcgcct 2160 ctcaaacacg acgtcgaggc tgtggtgcag atgcgcgtcc
tgcatgaagg tagtgaagag 2220 gaaagtaagg gatgtgtttt gtgggccttc gtggctgcgc acacacctct
tatctccttc 2280 gcttgggtacc ttctcccttt tctgtcttca ccccccttc tcttctcagc ctctccctgg
2340 cgcggtggtg caacgatttc gttttattta cgtctgtgta gctcctctat tcaacggtgc 2400
gatgacgcta acgaagctgg cctgtattcg gctaaggcga aggcaaaaga ctaggagggg 2460 ggggggaagg
agacggcgtg accatcactg cgaagaaaca agccgaagaa aaggccccga 2520 acgcctgcat ttccgcgcgc
cctcgcccg cttcttctct tcttctgctc tctctctctc 2580 tctctctcgc tatcttctca acggagacat
gaaaggcgtt tgtaggaaa agaggggggg 2640 gggaagagtg ggacgacgcg ctgcgtcttt tgggcactgg
tcacgtgcgt caccctcttt 2700 ttttatctct attggcactg tcttgtttct tttccctttc ctatcatacg
cgtctcgcaa 2760 acgactccgc gctgagcagc catgtgctgc ggcgtggagg aagtacacag acatcacgga
2820 tgcatatgtg cgcgtccgtg tacgcgcttg tatggggctt ctaacagcgc ctgtgtgtgt 2880
ttgtgtgtgt gtgtgtgtgt gtgtctgtgt atttcgagcg tctgtatgct attctattaa 2940 gcaccgaaga
agagacacac acgacagcga aggagatggt gtcggctttt cggctaata 3000 ctcccttcca tagcttctct
gaaggaggct ctcttccaga ggaatagact gcagatgggg 3060 tccacgttta tctgaggagt caacggaaaa
aaaaaaaaa aaaaaaaaaa aaaaaaaaaa 3120 aaaaaaaact cgag 3134 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 2 <211> LENGTH: 546 <212> TYPE: PRT <213>
ORGANISM: Leishmania major <400> SEQUENCE: 2 Met Asp Ala Thr Glu Leu Lys Asn Lys
Gly Asn Glu Glu Phe Ser Ala 1 5 10 15 Gly Arg Tyr Val Glu Ala Val Asn Tyr Phe Ser
Lys Ala Ile Gln Leu 20 25 30 Asp Glu Gln Asn Ser Val Leu Tyr Ser Asn Arg Ser Ala
Cys Phe Ala 35 40 45 Ala Met Gln Lys Tyr Lys Asp Ala Leu Asp Asp Ala Lys Cys
Ile 50 55 60 Ser Ile Lys Pro Asn Trp Ala Lys Gly Tyr Val Arg Arg Gly Ala Ala 65 70
75 80 Leu His Gly Met Arg Arg Tyr Asp Asp Ala Ile Ala Ala Tyr Glu Lys 85 90 95 Gly
Leu Lys Val Asp Pro Ser Asn Ser Gly Cys Ala Gln Gly Val Lys 100 105 110 Asp Val Gln
Val Ala Lys Ala Arg Glu Ala Arg Asp Pro Ile Ala Arg 115 120 125 Val Phe Thr Pro Glu
Ala Phe Arg Lys Ile Gln Glu Asn Pro Lys Leu 130 135 140 Ser Leu Leu Met Leu Gln Pro
Asp Tyr Val Lys Met Val Asp Thr Val 145 150 155 160 Ile Arg Asp Pro Ser Gln Gly Arg
Leu Tyr Met Glu Asp Gln Arg Phe 165 170 175 Ala Leu Thr Leu Met Tyr Leu Ser Gly Met
Lys Ile Pro Asn Asp Gly 180 185 190 Asp Gly Glu Glu Glu Glu Arg Pro Ser Ala Lys Ala
Ala Glu Thr Ala 195 200 205 Lys Pro Lys Glu Glu Lys Pro Leu Thr Asp Asn Glu Lys Glu
Ala Leu 210 215 220 Ala Leu Lys Glu Glu Gly Asn Lys Leu Tyr Leu Ser Lys Lys Phe Glu
225 230 235 240 Glu Ala Leu Thr Lys Tyr Gln Glu Ala Gln Val Lys Asp Pro Asn Asn 245
250 255 Thr Leu Tyr Ile Leu Asn Val Ser Ala Val Tyr Phe Glu Gln Gly Asp 260 265 270
Tyr Asp Lys Cys Ile Ala Glu Cys Glu His Gly Ile Glu His Gly Arg 275 280 285 Glu Asn
His Cys Asp Tyr Thr Ile Ile Ala Lys Leu Met Thr Arg Asn 290 295 300 Ala Leu Cys Leu
Gln Arg Gln Arg Lys Tyr Glu Ala Ala Ile Asp Leu 305 310 315 320 Tyr Lys Arg Ala Leu
Val Glu Trp Arg Asn Pro Asp Thr Leu Lys Lys 325 330 335 Leu Thr Glu Cys Glu Lys Glu
His Gln Lys Ala Val Glu Glu Ala Tyr 340 345 350 Ile Asp Pro Glu Ile Ala Lys Gln Lys
Lys Asp Glu Gly Asn Gln Tyr 355 360 365 Phe Lys Glu Asp Lys Phe Pro Glu Ala Val Ala
Ala Tyr Thr Glu Ala 370 375 380 Ile Lys Arg Asn Pro Ala Glu His Thr Ser Tyr Ser Asn
Arg Ala Ala 385 390 395 400 Ala Tyr Ile Lys Leu Gly Ala Phe Asn Asp Ala Leu Lys Asp
Ala Glu 405 410 415 Lys Cys Ile Glu Leu Lys Pro Asp Phe Val Lys Gly Tyr Ala Arg Lys
420 425 430 Gly His Ala Tyr Phe Trp Thr Lys Gln Tyr Asn Arg Ala Leu Gln Ala 435 440
445 Tyr Asp Glu Gly Leu Lys Val Asp Pro Ser Asn Ala Asp Cys Lys Asp 450 455 460 Gly
Arg Tyr Arg Thr Ile Met Lys Ile Gln Glu Met Ala Ser Gly Gln 465 470 475 480 Ser Ala
Asp Gly Asp Glu Ala Ala Arg Arg Ala Met Asp Asp Pro Glu 485 490 495 Ile Ala Ala Ile
Met Gln Asp Ser Tyr Met Gln Leu Val Leu Lys Glu 500 505 510 Met Gln Asn Asp Pro Thr
Arg Ile Gln Glu Tyr Met Lys Asp Ser Gly 515 520 525 Ile Ser Ser Lys Ile Asn Lys Leu
Ile Ser Ala Gly Ile Ile Arg Phe 530 535 540 Gly Gln 545 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 3 <211> LENGTH: 676 <212> TYPE: DNA <213>
ORGANISM: Leishmania donovani <400> SEQUENCE: 3 aattcggcac gaggcattgt gcataatggt
caagtcccac tacatctgcg cgggccgcct 60 ggtgcgcata ctgcgtggcc cccgccagga ccgcgttggt
gtgatcgctg acattgtcga 120 cgcaaccgc gtgctggtgg agaaccggga ggacgcgaag atgtggcgcc
acgtgcagaa 180 cctgaagaac gtggagccgc tgaagtactg cgtgagcgtc agccgcaact gcagcgcgaa
240 ggcgctgaag gatgcgtggt cctcgtcgaa ggcgctggag aagtacgcga agacgcgcac 300
tgctgcgcgc gtggaggcga agaaggcgtg cgccgcgtcg acggacttcg agcgtacca 360 gctgcgcgtt
gcgcgccggt ctgcgcgca ctgggcgcgc aggtgttctc acgaagga 420 cgcaagacg cccgtgtcgt
ggcacaagggt tgcgctgaag aagatgcga agaaggccgc 480 aaagatggac tcgaccgagg gcgctaagag
gcgcatgcag aaggcgatcg ctgcccga 540 ggcaaaaaag taaggccata ccctcacttc gcttgtttcg
tgatttttcg tgggagtcgg 600 tggccctacc agcggtcttt cattggctta tttctatccg gtctgaaaga
ggtacaaaaa 660 aaaaaaaaaa aaaaaa 676 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID
NO 4 <211> LENGTH: 175 <212> TYPE: PRT <213> ORGANISM: Leishmania donovani <400>

SEQUENCE: 4 Met Val Lys Ser His Tyr Ile Cys Ala Gly Arg Leu Val Arg Ile Leu 1 5 10
 15 Arg Gly Pro Arg Gln Asp Arg Val Gly Val Ile Val Asp Ile Val Asp 20 25 30 Ala Asn
 Arg Val Leu Val Glu Asn Pro Glu Asp Ala Lys Met Trp Arg 35 40 45 His Val Gln Asn
 Leu Lys Asn Val Glu Pro Leu Lys Tyr Cys Val Ser 50 55 60 Val Ser Arg Asn Cys Ser
 Ala Lys Ala Leu Lys Asp Ala Leu Ala Ser 65 70 75 80 Ser Lys Ala Leu Glu Lys Tyr Ala
 Lys Thr Arg Thr Ala Ala Arg Val 85 90 95 Glu Ala Lys Lys Ala Cys Ala Ala Ser Thr
 Asp Phe Glu Arg Tyr Gln 100 105 110 Leu Arg Val Ala Arg Arg Ser Arg Ala His Trp Ala
 Arg Lys Val Phe 115 120 125 Asp Glu Lys Asp Ala Lys Thr Pro Val Ser Trp His Lys Val
 Ala Leu 130 135 140 Lys Lys Met Gln Lys Lys Ala Ala Lys Met Asp Ser Thr Glu Gly Ala
 145 150 155 160 Lys Arg Arg Met Gln Lys Ala Ile Ala Ala Arg Lys Ala Lys Lys 165 170
 175 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 5 <211> LENGTH: 2040 <212>

TYPE: DNA <213> ORGANISM: Leishmania braziliensis <400> SEQUENCE: 5 cgcggtggcg
 gccgctctag aactagtggg tccccggggc tgcagggaatt cggcacgaga 60 gagcctgacg gacccggcgg
 tgctgggcca ggagactcac ctgcgcgtcc gcgtggtgcc 120 ggacaaggcg aacaagacgc tgacggtgga
 ggataacggc atcggcatga ccaaggcgga 180 cctcgtgaac aatctgggca cgcacgcgcg ctccggcacg
 aaggctttca tggaggcact 240 ggaggccggc ggcgacatga gcatgatcgg ccagttcggg gtcggcttct
 actccgcgta 300 ccttgtggcg gaccgcgtga cgggtggtgc gaagaacaac tcggacgagg cgtactggga
 360 atcgtctcgc gggggcacgt tcaccatcac gagcgtgcag gagtcggaca tgaagcgcg 420
 cacgagtaca acgctgcacc taaaggagga ccagcaggag tacctggagg agcgccgggt 480 gaaggagctg
 atcaagaagc actccgagtt catcggtac gacatcgagc tgatggtgga 540 gaagacggcg gagaaggagg
 tgacggacga ggacgaggag gaggacgagt cgaagaagaa 600 gtcctgcggg gacgaggcg agccgaaggt
 ggaggaggtg acggaaggcg gcgaggacaa 660 gaagaagaag acgaagaagg tgaaggaggt gaagaagacg
 tacgaggtga agaacaagca 720 caagccgctc tggacgcgcg acacgaagga cgtgacgaag gaggagtacg
 cggccttcta 780 caaggccatc tccaacgact gggaggacac ggcggcgacg aagcacttct cggtgagggg
 840 ccagctggag ttccgcgcga tcgcgttcgt gccgaagcgc gcgccgttcg acatgttcga 900
 gccgaacaag aagcgcaaca acatcaagct gtacgtgcgc cgcgtgttca tcatggacaa 960 ctgcgaggac
 ctgtgcccgg actggctcgg cttcgtgaag ggcgtcgtgg acagcgagga 1020 cctgccgctg aacatctcgc
 gcgagaacct gcagcagaac aagatcctga aggtgatccg 1080 caagaacatc gtgaagaagt gcctggagct
 gttcgaagag atagcggaga acaaggagga 1140 ctacaagcag ttctacgagc agttcggcaa gaacatcaag
 ctgggcatcc acgaggacac 1200 ggcgaaccgc aagaagctga tggagtgtgct gcgcttctac agcaccgagt
 cgggggagga 1260 gatgacgaca ctgaaggact acgtgacgcg catgaagccg gagcagaagt cgcactacta
 1320 catcactggc gacagcaaga agaagctgga gtcgtcgccg ttcacgaga aggcgagacg 1380
 ctgcgggctc gaggtgctgt tcatgacgga gccgatcgac gagtacgtga tgcagcaggt 1440 gaaggacttc
 gaggacaaga agttcgcgtg cctgacgaag gaaggcgtgc acttcgagga 1500 gtccgaggag gagaagaagc
 agcgcgagga gaagaaggcg gcgtgcgaga agctgtgcaa 1560 gacgatgaag gaggtgctgg gcgacaaggt
 ggagaaggtg accgtgtcgg agcgctgtt 1620 gacgtcgccg tgcacctcgg tgacgtcgga gtttgggtgg
 tcggcgaca tggaacagat 1680 catgcgcaac caggcgtgc gcgactccag catggcgag tacatggtgt
 ccaagaagac 1740 gatggaggtg aaccccgacc accccatcat caaggagctg cgcgcgcg tgaggcgga
 1800 cgagaacgac aaggccgtga aggacctcgt cttcctgctc ttcgacacgt cgctgctcac 1860
 gtccggcttc cagctggatg accccaccgg ctacgcgag cgcataacc gcatgatcaa 1920 gctcggcctg
 tcgctcgacg aggaggagga ggaggtcgcc gaggcggcg cgccgagggc 1980 agccccgcg gaggtcaccg
 ccggcacctc cagcatggag gaggtggact gagccggtaa 2040 <200> SEQUENCE CHARACTERISTICS:

<210> SEQ ID NO 6 <211> LENGTH: 656 <212> TYPE: PRT <213> ORGANISM: Leshmania
 brailiensis <400> SEQUENCE: 6 Ser Leu Thr Asp Pro Ala Val Leu Gly Glu Glu Thr His
 Leu Arg Val 1 5 10 15 Arg Val Val Pro Asp Lys Ala Asn Lys Thr Leu Thr Val Glu Asp
 Asn 20 25 30 Gly Ile Gly Met Thr Lys Ala Asp Leu Val Asn Asn Leu Gly Thr Ile 35 40
 45 Ala Arg Ser Gly Thr Lys Ala Phe Met Glu Ala Leu Glu Ala Gly Gly 50 55 60 Asp Met
 Ser Met Ile Gly Gln Phe Gly Val Gly Phe Tyr Ser Ala Tyr 65 70 75 80 Leu Val Ala Asp
 Arg Val Thr Val Val Ser Lys Asn Asn Ser Asp Glu 85 90 95 Ala Tyr Trp Glu Ser Ser
 Ala Gly Gly Thr Phe Thr Ile Thr Ser Val 100 105 110 Gln Glu Ser Asp Met Lys Arg Gly
 Thr Ser Thr Thr Leu His Leu Lys 115 120 125 Glu Asp Gln Gln Glu Tyr Leu Glu Glu Arg
 Arg Val Lys Glu Leu Ile

Detailed Description Paragraph Table (4):

130 135 140 Lys Lys His Ser Glu Phe Ile Gly Tyr Asp Ile Glu Leu Met Val Glu 145 150
 155 160 Lys Thr Ala Glu Lys Glu Val Thr Asp Glu Asp Glu Glu Asp Glu 165 170 175
 Ser Lys Lys Lys Ser Cys Gly Asp Glu Gly Glu Pro Lys Val Glu Glu 180 185 190 Val Thr
 Glu Gly Gly Glu Asp Lys Lys Lys Lys Thr Lys Lys Val Lys 195 200 205 Glu Val Lys Lys
 Thr Tyr Glu Val Lys Asn Lys His Lys Pro Leu Trp 210 215 220 Thr Arg Asp Thr Lys Asp
 Val Thr Lys Glu Glu Tyr Ala Ala Phe Tyr 225 230 235 240 Lys Ala Ile Ser Asn Asp Trp
 Glu Asp Thr Ala Ala Thr Lys His Phe 245 250 255 Ser Val Glu Gly Gln Leu Glu Phe Arg
 Ala Ile Ala Phe Val Pro Lys 260 265 270 Arg Ala Pro Phe Asp Met Phe Glu Pro Asn Lys
 Lys Arg Asn Asn Ile 275 280 285 Lys Leu Tyr Val Arg Arg Val Phe Ile Met Asp Asn Cys
 Glu Asp Leu 290 295 300 Cys Pro Asp Trp Leu Gly Phe Val Lys Gly Val Val Asp Ser Glu

h e b b g e e f c e

e ge

Asp 305 310 315 320 Leu Pro Leu Asn Ile Ser Arg Glu Asn Leu Gln Gln Asn Lys Ile Leu
325 330 335 Lys Val Ile Arg Lys Asn Ile Val Lys Lys Cys Leu Glu Leu Phe Glu 340 345
350 Glu Ile Ala Glu Asn Lys Glu Asp Tyr Lys Gln Phe Tyr Glu Gln Phe 355 360 365 Gly
Lys Asn Ile Lys Leu Gly Ile His Glu Asp Thr Ala Asn Arg Lys 370 375 380 Lys Leu Met
Glu Leu Leu Arg Phe Tyr Ser Thr Glu Ser Gly Glu Glu 385 390 395 400 Met Thr Thr Leu
Lys Asp Tyr Val Thr Arg Met Lys Pro Glu Gln Lys 405 410 415 Ser Ile Tyr Tyr Ile Thr
Gly Asp Ser Lys Lys Lys Leu Glu Ser Ser 420 425 430 Pro Phe Ile Glu Lys Ala Arg Arg
Cys Gly Leu Glu Val Leu Phe Met 435 440 445 Thr Glu Pro Ile Asp Glu Tyr Val Met Gln
Gln Val Lys Asp Phe Glu 450 455 460 Asp Lys Lys Phe Ala Cys Leu Thr Lys Glu Gly Val
His Phe Glu Glu 465 470 475 480 Ser Glu Glu Glu Lys Lys Gln Arg Glu Glu Lys Lys Ala
Ala Cys Glu 485 490 495 Lys Leu Cys Lys Thr Met Lys Glu Val Leu Gly Asp Lys Val Glu
Lys 500 505 510 Val Thr Val Ser Glu Arg Leu Leu Thr Ser Pro Cys Ile Leu Val Thr 515
520 525 Ser Glu Phe Gly Trp Ser Ala His Met Glu Gln Ile Met Arg Asn Gln 530 535 540
Ala Leu Arg Asp Ser Ser Met Ala Gln Tyr Met Val Ser Lys Lys Thr 545 550 555 560 Met
Glu Val Asn Pro Asp His Pro Ile Ile Lys Glu Leu Arg Arg Arg 565 570 575 Val Glu Ala
Asp Glu Asn Asp Lys Ala Val Lys Asp Leu Val Phe Leu 580 585 590 Leu Phe Asp Thr Ser
Leu Leu Thr Ser Gly Phe Gln Leu Asp Asp Pro 595 600 605 Thr Gly Tyr Ala Glu Arg Ile
Asn Arg Met Ile Lys Leu Gly Leu Ser 610 615 620 Leu Asp Glu Glu Glu Glu Glu Val Ala
Glu Ala Pro Pro Ala Glu Ala 625 630 635 640 Ala Pro Ala Glu Val Thr Ala Gly Thr Ser
Ser Met Glu Gln Val Asp 645 650 655 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO
7 <211> LENGTH: 1771 <212> TYPE: DNA <213> ORGANISM: Leishmania tropica <400>
SEQUENCE: 7 caggcccgcg tccaggccct cgaggaggca gcgcgtctcc gcgcggagct ggaggcggcc 60
gaggaggcgg cccgcctgga tgtcatgcat gcggccgagc aggcccggtgt ccaggccctc 120 gaggaggcag
cgcgctctccg cgccggagctg gaggaggccg aggaggcgcc cgccctggat 180 gtcatgcatg cgcccgagca
ggcccgcgtc caggccctcg aggaggcagc gcgtctccgc 240 gcggagctgg aggctgccga ggaggcgggc
cgcttgagg ccatgcacga ggccgagcag 300 gcccgctccc aggccctcga ggaggcagcg gctctccgcg
cgagctgga ggaagccgag 360 gaggcggccc gcctggatgt catgcatgcg gccgagcagg cccgcgtcca
ggccctcgag 420 gaggcagcgc gtctccgcgc ggagctggag gaggcgagg aggcggcccc cctggaggcc
480 atgcacgagg ccgagcaggc ccgctcccag gccctcgagg aggcagcgcg tctccgcgcg 540
gagctggagg cggccgagga ggcggccgcg ctggatgtca tgcacgaggc cgagcaggcc 600 cgtgtccagg
ccctcgagga ggcggcgcg ctggatgtca tgcacgaggc cgagcaggcc 660 cgcgctccagg ccctcgagga
ggcagcgcgt ctccgcgcg agctggaggc ggccgaggag 720 gcggcccgcc tggatgtcat gcacgaggcc
gagcaggccc gcgtccaggc cctcgaggag 780 gcagcgcgtc tccgcgcgga gctggaggcg gccgaggagg
cgccccgcct ggatgtcatg 840 cacgaggggc agcaggcccc tgtccaggcc ctcgaggagg cggccgcct
ggaggccatg 900 cacgaggccg agcaggcccc ctcccaggcc ctcgaggagg cagcgcgctt ctgcgcggag
960 ctggaggctg aggaggagga aaaagatgag cggccggcga cgtcgagcta cagcgaggag 1020
tgcaaagggc gactgctatc gagggcgcg ccgcatccgc ggaggccgct gccgcggccg 1080 ttcatggga
tgtactgtt ggaggatgtg gagaagagta ttctcattgt ggacgggctc 1140 tacagggatg ggccggcgta
ccagacgggc atccgcctcg gggatgtcct ctgtcgatc 1200 gcgggggttt acgtggattc aatagcgaag
gcgaggcagg tggatgagc gcgttgcgc 1260 tgcggctgcg tcgttcccg gacgtggcg acgaagatga
accagcagta cagcgtggct 1320 ctgtatatca tgacggtgga tccgcagcac aacgacaagc cctttttttt
tgatgtgcac 1380 atccaccacc gcacgagag ctcgcacatg gggaagaagg cgcagtggat ggaagtctt
1440 gagagcccat ccgtatcttc ggctgccacc acccctctcg tgccgctctt gcgtgagccg 1500
acgcgcgta ggggctcaga gctgcagtca agtgctcgtt ccgccttcgt tgccacgtct 1560 tacttctcga
gcgcgcgag gtcggtcagc tcagaaagt agcagccgcg cgggtcctct 1620 agcgtggcta tggcgaggga
ggcgatcgcg ctggcgccgc aagggtatac cccacccaac 1680 caagtgcgcg gccgtagttg acgtctctgt
gtgagtgtgt gtcgctccgt ctcttctctt 1740 ttctgcatg tgttttattc atttcttttt c 1771 <200>
SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 8 <211> LENGTH: 566 <212> TYPE: PRT <213>
ORGANISM: Leishmania tropica <400> SEQUENCE: 8 Gln Ala Arg Val Gln Ala Leu Glu Glu
Ala Ala Arg Leu Arg Ala Glu 1 5 10 15 Leu Glu Ala Ala Glu Glu Ala Ala Arg Leu Asp
Val Met His Ala Ala 20 25 30 Glu Gln Ala Arg Val Gln Ala Leu Glu Glu Ala Ala Arg
Leu Arg Ala 35 40 45 Glu Leu Glu Glu Ala Glu Glu Ala Ala Arg Leu Asp Val Met His
Ala 50 55 60 Ala Glu Gln Ala Arg Val Gln Ala Leu Glu Glu Ala Ala Arg Leu Arg 65 70
75 80 Ala Glu Leu Glu Ala Ala Glu Glu Ala Ala Arg Leu Glu Ala Met His 85 90 95 Glu
Ala Glu Gln Ala Arg Ser Gln Ala Leu Glu Glu Ala Ala Arg Leu 100 105 110 Arg Ala Glu
Leu Glu Glu Ala Glu Glu Ala Ala Arg Leu Asp Val Met 115 120 125 His Ala Ala Glu Gln
Ala Arg Val Gln Ala Leu Glu Glu Ala Ala Arg 130 135 140 Leu Arg Ala Glu Leu Glu Glu
Ala Glu Glu Ala Ala Arg Leu Glu Ala 145 150 155 160 Met His Glu Ala Glu Gln Ala Arg
Ser Gln Ala Leu Glu Glu Ala Ala 165 170 175 Arg Leu Arg Ala Glu Leu Glu Ala Ala Glu
Glu Ala Ala Arg Leu Asp 180 185 190 Val Met His Glu Ala Glu Gln Ala Arg Val Gln Ala
Leu Glu Glu Ala 195 200 205 Ala Arg Leu Asp Val Met His Glu Ala Glu Gln Ala Arg Val
Gln Ala 210 215 220 Leu Glu Glu Ala Ala Arg Leu Arg Ala Glu Leu Glu Ala Ala Glu Glu
225 230 235 240 Ala Ala Arg Leu Asp Val Met His Glu Ala Glu Gln Ala Arg Val Gln 245
250 255 Ala Leu Glu Glu Ala Ala Arg Leu Arg Ala Glu Leu Glu Ala Ala Glu 260 265 270

Glu Ala Ala Arg Leu Asp Val Met His Glu Gly Glu Gln Ala Arg Val 275 280 285 Gln Ala
 Leu Glu Glu Ala Ala Arg Leu Glu Ala Met His Glu Ala Glu 290 295 300 Gln Ala Arg Ser
 Gln Ala Leu Glu Glu Ala Ala Arg Leu Cys Ala Glu 305 310 315 320 Leu Glu Ala Glu Glu
 Glu Glu Lys Asp Glu Arg Pro Ala Thr Ser Ser 325 330 335 Tyr Ser Glu Glu Cys Lys Gly
 Arg Leu Leu Ser Arg Ala Arg Pro Asp 340 345 350 Pro Arg Arg Pro Leu Pro Arg Pro Phe
 Ile Gly Met Ser Leu Leu Glu 355 360 365 Asp Val Glu Lys Ser Ile Leu Ile Val Asp Gly
 Leu Tyr Arg Asp Gly 370 375 380 Pro Ala Tyr Gln Thr Gly Ile Arg Leu Gly Asp Val Leu
 Leu Arg Ile 385 390 395 400 Ala Gly Val Tyr Val Asp Ser Ile Ala Lys Ala Arg Gln Val
 Val Asp 405 410 415 Ala Arg Cys Arg Cys Gly Cys Val Val Pro Val Thr Leu Ala Thr Lys
 420 425 430 Met Asn Gln Gln Tyr Ser Val Ala Leu Tyr Ile Met Thr Val Asp Pro 435 440
 445 Gln His Asn Asp Lys Pro Phe Phe Phe Asp Val His Ile His His Arg 450 455 460 Ile
 Glu Ser Ser His Met Gly Lys Lys Ala Gln Trp Met Glu Val Leu 465 470 475 480 Glu Ser
 Pro Ser Val Ser Ser Ala Ala Thr Thr Pro Leu Val Pro Leu 485 490 495 Leu Arg Glu Pro
 Thr Pro Arg Arg Gly Ser Glu Leu Gln Ser Ser Ala 500 505 510 Arg Ser Ala Phe Val Ala
 Thr Ser Tyr Phe Ser Ser Ala Arg Arg Ser 515 520 525 Val Ser Ser Glu Ser Glu Arg Pro
 Arg Gly Ser Ser Ser Val Ala Met 530 535 540 Ala Glu Glu Ala Ile Ala Leu Ala Pro Gln
 Gly Tyr Thr Pro Pro Asn 545 550 555 560 Gln Val Arg Gly Arg Ser 565 <200> SEQUENCE

CHARACTERISTICS: <210> SEQ ID NO 9 <211> LENGTH: 1618 <212> TYPE: DNA <213>

ORGANISM: *Leishmania braziliensis* <400> SEQUENCE: 9 ccactctctc ggctcgctctgt
 ctccacgcgc cgcacgcagt tgatttccgc ctctcttaaac 60 gctctctttt tttttatttt tcacctgacc
 aaccgcacca cgtcggcctc catcatgtcg 120 cagcaagacc gagttgcccc acaggaccag gactcgttcc
 tcgacgacca gcccggcgtc 180 cgcccgatcc cgtccttcga tgacatgccg ttgcaccaga accttctgcg
 cggcatctac 240 tcgtacggct tcgagaaacc gtccagcatc cagcagcgcg ccacgcgcgc cttcacgcgc
 300 ggcggcgaca tcacgcgcga ggcgcagtc ccgtaccggca agacgggcgc cttctccatc 360
 ggctgctgc agcgctgga ctccgcac aacctgatcc agggcctcgt gctctccccg 420 accgcgcgagc
 tggccctgca cagcgcgag gtgatcagcc gcacgcgca gttcctgtcg 480 aacagcgca agttctgtga
 gacctttgtg ggtggcacgc gcgtgcagga tgacctgcgc 540 aagctgcagg ctggcgctcg cgtcgccgtg
 gggacgcccgc gccgcgtgtc cgacgtgatc 600 aagcgcgcg cgctgcgcac cgagtccttg cgcgtgctgg
 tgctcgacga ggctgatgag 660 atgctgtctc agggcttcgc ggatcagatt tacgagatct tccgcttcct
 gccgaaggac 720 atccaggtcg cgctcttctc cgccacgatg ccggaggagg tgctggagct gacaaagaag
 780 ttcacgcgcg acccgtacg cattctcgtg aagcgcgaga gcctgacgct ggagggcac 840
 aagcagttct tcacgcgcgt cgaggaggag cacaagctgg acacgctgat ggacctgtac 900 gagaccgtgt
 ccacgcgcga gtccgtcatc ttccgcaaca cccgcgcga ggtggactgg 960 atcgccgaga agctgaatca
 gagcaaccac accgtcagca gcacgcacgc cgagatgccc 1020 aagagcgacc gcgagcgct catgaacacc
 ttccgcagcg gcagctcccgc cgtgctcgta 1080 acgaccgacc tcgtggcccc cgcatcgac gtgcaccacg
 tgaacatcgt catcaacttc 1140 gacctgccga cgaacaagga gaactacctg caccgcattg gccgcggcgg
 ccgctacggc 1200 gtaaagggtg ttgccatcaa ctctgtgacg gagaaagacg tggagctgct gcacgagatc
 1260 gaggggcact accacacgca gatcgatgag ctcccggtgg actttgccgc ctacctcggc 1320
 gactgagcgg gccctgccc ccttccctg cccccctctc gcgacgagag aacgcacatc 1380 gtaacacagc
 cagcgaaacg atagtaaggc cgtgcggcgc cgttccctc ctctgccag 1440 cgccccct ccgcagcgt
 tctcttttga gaggggggca gggggaggcg ctgcgcctgg 1500 ctggatgtgt gcttgagctt gcattccgtc
 aagcaagtgc ttgttttaa ttatgcgcgc 1560 cgttttgttg ctgctccctt tcgttggtgt ttttccggcc
 gaaacggcgt ttaagca 1618 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 10 <211>
 LENGTH: 403 <212> TYPE: PRT <213> ORGANISM: *Leishmania braziliensis* <400> SEQUENCE:

10. Met Ser Gln Gln Asp Arg Val Ala Pro Gln Asp Gln Asp Ser Phe Leu 1 5 10 15 Asp
 Asp Gln Pro Gly Val Arg Pro Ile Pro Ser Phe Asp Asp Met Pro 20 25 30 Leu His Gln
 Asn Leu Leu Arg Gly Ile Tyr Ser Tyr Gly Phe Glu Lys 35 40 45 Pro Ser Ser Ile Gln
 Gln Arg Ala Ile Ala Pro Phe Thr Arg Gly Gly 50 55 60 Asp Ile Ile Ala Gln Ala Gln
 Ser Gly Thr Gly Lys Thr Gly Ala Phe 65 70 75 80 Ser Ile Gly Leu Leu Gln Arg Leu Asp
 Phe Arg His Asn Leu Ile Gln 85 90 95 Gly Leu Val Leu Ser Pro Thr Arg Glu Leu Ala
 Leu Gln Thr Ala Glu 100 105 110 Val Ile Ser Arg Ile Gly Glu Phe Leu Ser Asn Ser Ala
 Lys Phe Cys 115 120 125 Glu Thr Phe Val Gly Gly Thr Arg Val Gln Asp Asp Leu Arg Lys
 Leu 130 135 140 Gln Ala Gly Val Val Val Ala Val Gly Thr Pro Gly Arg Val Ser Asp 145
 150 155 160 Val Ile Lys Arg Gly Ala Leu Arg Thr Glu Ser Leu Arg Val Leu Val 165 170
 175 Leu Asp Glu Ala Asp Glu Met Leu Ser Gln Gly Phe Ala Asp Gln Ile 180 185 190 Tyr
 Glu Ile Phe Arg Phe Leu Pro Lys Asp Ile Gln Val Ala Leu Phe 195 200 205 Ser Ala Thr
 Met Pro Glu Glu Val Leu Glu Leu Thr Lys Lys Phe Met 210 215 220 Arg Asp Pro Val Arg
 Ile Leu Val Lys Arg Glu Ser Leu Thr Leu Glu 225 230 235 240 Gly Ile Lys Gln Phe Phe
 Ile Ala Val Glu Glu Glu His Lys Leu Asp 245 250 255 Thr Leu Met Asp Leu Tyr Glu Thr
 Val Ser Ile Ala Gln Ser Val Ile

Detailed Description Paragraph Table (5):

260 265 270 Phe Ala Asn Thr Arg Arg Lys Val Asp Trp Ile Ala Glu Lys Leu Asn 275 280
 285 Gln Ser Asn His Thr Val Ser Ser Met His Ala Glu Met Pro Lys Ser 290 295 300 Asp

h e b b g e e f c e

e ge

Arg Glu Arg Val Met Asn Thr Phe Arg Ser Gly Ser Ser Arg Val 305 310 315 320 Leu Val
Thr Thr Asp Leu Val Ala Arg Gly Ile Asp Val His His Val 325 330 335 Asn Ile Val Ile
Asn Phe Asp Leu Pro Thr Asn Lys Glu Asn Tyr Leu 340 345 350 His Arg Ile Gly Arg Gly
Gly Arg Tyr Gly Val Lys Gly Val Ala Ile 355 360 365 Asn Phe Val Thr Glu Lys Asp Val
Glu Leu Leu His Glu Ile Glu Gly 370 375 380 His Tyr His Thr Gln Ile Asp Glu Leu Pro
Val Asp Phe Ala Ala Tyr 385 390 395 400 Leu Gly Glu <200> SEQUENCE CHARACTERISTICS:
<210> SEQ ID NO 11 <211> LENGTH: 12 <212> TYPE: PRT <213> ORGANISM: Leishmania
donovani <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION: (1)...(5) <223>
OTHER INFORMATION: Xaa = any amino acid <400> SEQUENCE: 11 Xaa Gln Xaa Pro Gln Xaa
Val Phe Asp Glu Xaa Xaa 1 5 10 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 12
<211> LENGTH: 26 <212> TYPE: DNA <213> ORGANISM: Artificial Sequence <220> FEATURE:
<223> OTHER INFORMATION: Sense PCR pri mer <400> SEQUENCE: 12 ggaattcccc ncagctngtn
ttcgac 26 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 13 <211> LENGTH: 5 <212>
TYPE: PRT <213> ORGANISM: Leishmania donovani <400> SEQUENCE: 13 Lys Val Phe Asp
Glu 1 5 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 14 <211> LENGTH: 30 <212>
TYPE: DNA <213> ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER
INFORMATION: PCR primer <400> SEQUENCE: 14 ggatccatgg tcaagtccca ctacatctgc 30
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 15 <211> LENGTH: 33 <212> TYPE: DNA
<213> ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR
primer <400> SEQUENCE: 15 gaattcagac cggatagaaa taagccaatg aaa 33 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 16 <211> LENGTH: 701 <212> TYPE: PRT <213>
ORGANISM: Leishmania amazonensis <400> SEQUENCE: 16 Met Thr Glu Thr Phe Ala Phe Gln
Ala Glu Ile Asn Gln Leu Met Ser 1 5 10 15 Leu lle Ile Asn Thr Phe Tyr Ser Asn Lys
Glu Ile Phe Leu Arg Asp 20 25 30 Val Ile Ser Asn Ala Ser Asp Ala Cys Asp Lys Ile
Arg Tyr Gln Ser 35 40 45 Leu Thr Asp Pro Ala Val Leu Gly Asp Ala Thr Arg Leu Cys
Val Arg 50 55 60 Val Val Pro Asp Lys Glu Asn Lys Thr Leu Thr Val Glu Asp Asn Gly 65
70 75 80 Ile Gly Met Thr Lys Ala Asp Leu Val Asn Asn Leu Gly Thr Ile Ala 85 90 95
Arg Ser Gly Thr Lys Ala Phe Met Glu Ala Leu Glu Ala Gly Ala Asp 100 105 110 Met Ser
Met Ile Gly Gln Phe Gly Val Gly Phe Tyr Ser Ala Tyr Leu 115 120 125 Val Ala Asp Arg
Val Thr Val Thr Ser Lys Asn Asn Ser Asp Glu Val 130 135 140 Tyr Val Trp Glu Ser Ser
Ala Gly Gly Thr Phe Thr Ile Thr Ser Ala 145 150 155 160 Pro Glu Ser Asp Met Lys Leu
Pro Ala Arg Ile Thr Leu His Leu Lys 165 170 175 Glu Asp Gln Leu Glu Tyr Leu Glu Ala
Arg Arg Leu Lys Glu Leu Ile 180 185 190 Lys Lys His Ser Glu Phe Ile Gly Tyr Asp Ile
Glu Leu Met Val Glu 195 200 205 Lys Thr Thr Glu Lys Glu Val Thr Asp Glu Asp Glu Glu
Glu Ala Lys 210 215 220 Lys Ala Asp Glu Asp Gly Glu Glu Pro Lys Val Glu Glu Val Thr
Glu 225 230 235 240 Gly Glu Glu Asp Lys Lys Lys Lys Thr Lys Lys Val Lys Glu Val Thr
245 250 255 Lys Glu Tyr Glu Val Gln Asn Lys His Lys Pro Leu Trp Thr Arg Asp 260 265
270 Pro Lys Asp Val Thr Lys Glu Glu Tyr Ala Ala Phe Tyr Lys Ala Ile 275 280 285 Ser
Asn Asp Trp Glu Asp Pro Pro Ala Thr Lys His Phe Ser Val Glu 290 295 300 Gly Gln Leu
Glu Phe Arg Ala Ile Met Phe Val Pro Lys Arg Ala Pro 305 310 315 320 Phe Asp Met Leu
Glu Pro Asn Lys Lys Arg Asn Asn Ile Lys Leu Tyr 325 330 335 Val Arg Arg Val Phe Ile
Met Asp Asn Cys Glu Asp Leu Cys Pro Asp 340 345 350 Trp Leu Gly Phe Val Lys Gly Val
Val Asp Ser Glu Asp Leu Pro Leu 355 360 365 Asn Ile Ser Arg Glu Asn Leu Gln Gln Asn
Lys Ile Leu Lys Val Ile 370 375 380 Arg Lys Asn Ile Val Lys Lys Cys Leu Glu Met Phe
Glu Glu Val Ala 385 390 395 400 Glu Asn Lys Glu Asp Tyr Lys Gln Phe Tyr Glu Gln Phe
Gly Lys Asn 405 410 415 Ile Lys Leu Gly Ile His Glu Asp Thr Ala Asn Arg Lys Lys Leu
Met 420 425 430 Glu Leu Leu Arg Phe Tyr Ser Thr Glu Ser Gly Glu Val Met Thr Thr 435
440 445 Leu Lys Asp Tyr Val Thr Arg Met Lys Ala Glu Gln Asn Ser Ile Tyr 450 455 460
Tyr Ile Thr Gly Asp Ser Lys Lys Lys Leu Glu Ser Ser Pro Phe Ile 465 470 475 480 Glu
Gln Ala Lys Arg Arg Gly Phe Glu Val Leu Phe Met Thr Glu Pro 485 490 495 Tyr Asp Glu
Tyr Val Met Gln Gln Val Lys Asp Phe Glu Asp Lys Lys 500 505 510 Phe Ala Cys Leu Thr
Lys Glu Gly Val His Phe Glu Glu Ser Glu Glu 515 520 525 Glu Lys Lys Gln Arg Glu Glu
Glu Lys Ala Thr Cys Glu Lys Leu Cys 530 535 540 Lys Thr Met Lys Glu Val Leu Gly Asp
Lys Val Glu Lys Val Thr Val 545 550 555 560 Ser Glu Arg Leu Ser Thr Ser Pro Cys Ile
Leu Val Thr Ser Glu Phe 565 570 575 Gly Trp Ser Ala His Met Glu Gln Met Met Arg Asn
Gln Ala Leu Arg 580 585 590 Asp Ser Ser Met Ala Gln Tyr Met Met Ser Lys Lys Thr Met
Glu Leu 595 600 605 Asn Pro Lys His Pro Ile Ile Lys Glu Leu Arg Arg Arg Val Glu Ala
610 615 620 Asp Glu Asn Asp Lys Ala Val Lys Asp Leu Val Phe Leu Leu Phe Asp 625 630
635 640 Thr Ser Leu Leu Thr Ser Gly Phe Gln Leu Glu Asp Pro Thr Tyr Ala 645 650 655
Glu Arg Ile Asn Arg Met Ile Lys Leu Gly Leu Ser Leu Asp Glu Glu 660 665 670 Glu Glu
Glu Glu Ala Val Glu Ala Ala Val Ala Glu Thr Ala Pro Ala 675 680 685 Glu Val Thr Ala
Gly Thr Ser Ser Met Glu Leu Val Asp 690 695 700 <200> SEQUENCE CHARACTERISTICS:
<210> SEQ ID NO 17 <211> LENGTH: 704 <212> TYPE: PRT <213> ORGANISM: T. Cruzi <400>
SEQUENCE: 17 Met Thr Glu Thr Phe Ala Phe Gln Ala Glu Ile Asn Gln Leu Met Ser 1 5 10

15 Leu Ile Ile Asn Thr Phe Tyr Ser Asn Lys Glu Ile Phe Leu Arg Glu 20 25 30 Leu Ile
 Ser Asn Ala Ser Asp Ala Cys Asp Lys Ile Arg Tyr Gln Ser 35 40 45 Leu Thr Asn Gln
 Ala Val Leu Gly Asp Glu Ser His Leu Arg Ile Arg 50 55 60 Val Val Pro Asp Lys Ala
 Asn Lys Thr Leu Thr Val Glu Asp Thr Gly 65 70 75 80 Ile Gly Met Thr Lys Ala Glu Leu
 Val Asn Asn Leu Gly Thr Ile Ala 85 90 95 Arg Ser Gly Thr Lys Ala Phe Met Glu Ala
 Leu Glu Ala Gly Gly Asp 100 105 110 Met Ser Met Ile Gly Gln Phe Gly Val Gly Phe Tyr
 Ser Ala Tyr Leu 115 120 125 Val Ala Asp Arg Val Thr Val Val Ser Lys Asn Asn Asp Asp
 Glu Ala 130 135 140 Tyr Thr Trp Glu Ser Ser Ala Gly Gly Thr Phe Thr Val Thr Pro Thr
 145 150 155 160 Pro Asp Cys Asp Leu Lys Arg Gly Thr Arg Ile Val Leu His Leu Lys 165
 170 175 Glu Asp Gln Gln Glu Tyr Leu Glu Glu Arg Arg Leu Lys Asp Leu Ile 180 185 190
 Lys Lys His Ser Glu Phe Ile Gly Tyr Asp Ile Glu Leu Met Val Glu 195 200 205 Lys Ala
 Thr Glu Lys Glu Val Thr Asp Glu Asp Glu Asp Glu Ala Ala 210 215 220 Ala Thr Lys Asn
 Glu Glu Gly Glu Glu Pro Lys Val Glu Glu Val Lys 225 230 235 240 Asp Asp Ala Glu Glu
 Gly Glu Lys Lys Lys Lys Thr Lys Lys Val Lys 245 250 255 Glu Val Thr Gln Glu Phe Val
 Val Gln Asn Lys His Lys Pro Leu Trp 260 265 270 Thr Arg Asp Pro Lys Asp Val Thr Lys
 Glu Glu Tyr Ala Ala Phe Tyr 275 280 285 Lys Ala Ile Ser Asn Asp Trp Glu Glu Pro Leu
 Ser Thr Lys His Phe 290 295 300 Ser Val Glu Gly Gln Leu Glu Phe Arg Ala Ile Leu Phe
 Val Pro Lys 305 310 315 320 Arg Ala Pro Phe Asp Met Phe Glu Pro Ser Lys Lys Arg Asn
 Asn Ile 325 330 335 Lys Leu Tyr Val Arg Arg Val Phe Ile Met Asp Asn Cys Glu Asp Leu
 340 345 350 Cys Pro Glu Trp Leu Ala Phe Val Arg Gly Val Val Asp Ser Glu Asp 355 360
 365 Leu Pro Leu Asn Ile Ser Arg Glu Asn Leu Gln Gln Asn Lys Ile Leu 370 375 380 Lys
 Val Ile Arg Lys Asn Ile Val Lys Lys Ala Leu Glu Leu Phe Glu 385 390 395 400 Glu Ile
 Ala Glu Asn Lys Glu Asp Tyr Lys Lys Phe Tyr Glu Gln Phe 405 410 415 Gly Lys Asn Val
 Lys Leu Gly Ile His Glu Asp Ser Ala Asn Arg Lys 420 425 430 Lys Leu Met Glu Leu Leu
 Arg Phe His Ser Ser Glu Ser Gly Glu Asp 435 440 445 Met Thr Thr Leu Lys Asp Tyr Val
 Thr Arg Met Lys Glu Gly Gln Lys 450 455 460 Cys Ile Tyr Tyr Val Thr Gly Asp Ser Lys
 Lys Lys Leu Glu Thr Ser 465 470 475 480 Pro Phe Ile Glu Gln Ala Arg Arg Arg Gly Phe
 Glu Val Leu Phe Met 485 490 495 Thr Glu Pro Ile Asp Glu Tyr Val Met Gln Gln Val Lys
 Asp Phe Glu 500 505 510 Asp Lys Lys Phe Ala Cys Leu Thr Lys Glu Gly Val His Phe Glu
 Glu 515 520 525 Thr Glu Glu Glu Lys Lys Gln Arg Glu Glu Glu Lys Thr Ala Tyr Glu 530
 535 540 Arg Leu Cys Lys Ala Met Lys Asp Val Leu Gly Asp Lys Val Glu Lys 545 550 555
 560 Val Val Val Ser Glu Arg Leu Ala Thr Ser Pro Cys Ile Leu Val Thr 565 570 575 Ser
 Glu Phe Gly Trp Ser Ala His Met Glu Gln Ile Met Arg Asn Gln 580 585 590 Ala Leu Arg
 Asp Ser Ser Met Ser Ala Tyr Met Met Ser Lys Lys Thr 595 600 605 Met Glu Ile Asn Pro
 Ala His Pro Ile Val Lys Glu Leu Lys Arg Arg 610 615 620 Val Glu Ala Asp Glu Asn Asp
 Lys Ala Val Lys Asp Leu Val Tyr Leu 625 630 635 640 Leu Phe Asp Thr Ala Leu Leu Thr
 Ser Gly Phe Thr Leu Asp Asp Pro 645 650 655 Thr Ser Tyr Ala Glu Arg Ile His Arg Met
 Ile Lys Leu Gly Leu Ser 660 665 670 Leu Asp Asp Glu Asp Asn Gly Asn Glu Glu Ala Glu
 Pro Ala Ala Ala 675 680 685

Detailed Description Paragraph Table (6):

Val Pro Ala Glu Pro Val Ala Gly Thr Ser Ser Met Glu Gln Val Asp 690 695 700 <200>
 SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 18 <211> LENGTH: 732 <212> TYPE: PRT
 <213> ORGANISM: Homo sapien <400> SEQUENCE: 18 Met Pro Glu Glu Thr Gln Thr Gln Asp
 Gln Pro Met Glu Glu Glu Glu 1 5 10 15 Val Glu Thr Phe Ala Phe Gln Ala Glu Ile Ala
 Gln Leu Met Ser Leu 20 25 30 Ile Ile Asn Thr Phe Tyr Ser Asn Lys Glu Ile Phe Leu
 Arg Glu Leu 35 40 45 Ile Ser Asn Ser Ser Asp Ala Leu Asp Lys Ile Arg Tyr Glu Ser
 Leu 50 55 60 Thr Asp Pro Ser Lys Leu Asp Ser Gly Lys Glu Leu His Ile Asn Leu 65 70
 75 80 Ile Pro Asn Lys Gln Asp Arg Ala Leu Thr Ile Val Asp Thr Gly Ile 85 90 95 Gly
 Met Thr Lys Ala Asp Leu Ile Asn Asn Leu Gly Thr Ile Ala Lys 100 105 110 Ser Gly Thr
 Lys Ala Phe Met Glu Ala Leu Gln Ala Gly Ala Asp Ile 115 120 125 Ser Met Ile Gly Gln
 Phe Gly Val Gly Phe Tyr Ser Ala Tyr Leu Val 130 135 140 Ala Glu Lys Val Thr Val Ile
 Thr Lys His Asn Asp Asp Glu Gln Tyr 145 150 155 160 Ala Trp Glu Ser Ser Ala Gly Gly
 Ser Phe Thr Val Arg Thr Asp Thr 165 170 175 Gly Glu Pro Met Gly Arg Gly Thr Lys Val
 Ile Leu His Leu Lys Glu 180 185 190 Asp Gln Thr Glu Tyr Leu Glu Glu Arg Arg Ile Lys
 Glu Ile Val Lys 195 200 205 Lys His Ser Gln Phe Ile Gly Tyr Pro Ile Thr Leu Phe Val
 Glu Lys 210 215 220 Glu Arg Asp Lys Glu Val Ser Asp Asp Glu Ala Glu Glu Lys Glu Asp
 225 230 235 240 Lys Glu Glu Glu Lys Glu Lys Glu Glu Lys Glu Ser Glu Asp Lys Pro 245
 250 255 Glu Ile Glu Asp Val Gly Ser Asp Glu Glu Asp Glu Lys Lys Asp Gly 260 265 270
 Asp Lys Lys Lys Lys Lys Lys Ile Lys Glu Lys Tyr Ile Asp Lys Glu 275 280 285 Glu Leu
 Asn Lys Thr Lys Pro Ile Trp Thr Arg Asn Pro Asp Asp Ile 290 295 300 Thr Asn Glu Glu
 Tyr Gly Glu Phe Tyr Lys Ser Leu Thr Asn Asp Trp 305 310 315 320 Glu Asp His Leu Ala
 Val Lys His Phe Ser Val Glu Gly Gln Leu Glu 325 330 335 Phe Arg Ala Leu Leu Phe Val
 Pro Arg Arg Ala Pro Phe Asp Leu Phe 340 345 350 Glu Asn Arg Lys Lys Lys Asn Asn Ile

Lys Leu Tyr Val Arg Arg Val 355 360 365 Phe Ile Met Asp Asn Cys Glu Glu Leu Ile Pro
Glu Tyr Leu Asn Phe 370 375 380 Ile Arg Gly Val Val Asp Ser Glu Asp Leu Pro Leu Asn
Ile Ser Arg 385 390 395 400 Glu Met Leu Gln Gln Ser Lys Ile Leu Lys Val Ile Arg Lys
Asn Leu 405 410 415 Val Lys Lys Cys Leu Glu Leu Phe Thr Glu Leu Ala Glu Asp Lys Glu
420 425 430 Asn Tyr Lys Lys Phe Tyr Glu Gln Phe Ser Lys Asn Ile Lys Leu Gly 435 440
445 Ile His Glu Asp Ser Gln Asn Arg Lys Lys Leu Ser Glu Leu Leu Arg 450 455 460 Tyr
Tyr Thr Ser Ala Ser Gly Asp Glu Met Val Ser Leu Lys Asp Tyr 465 470 475 480 Cys Thr
Arg Met Lys Glu Asn Gln Lys His Ile Tyr Tyr Ile Thr Gly 485 490 495 Glu Thr Lys Asp
Gln Val Ala Asn Ser Ala Phe Val Glu Arg Leu Arg 500 505 510 Lys His Gly Leu Glu Val
Ile Tyr Met Ile Glu Pro Ile Asp Glu Tyr 515 520 525 Cys Val Gln Gln Leu Lys Glu Phe
Glu Gly Lys Thr Leu Val Ser Val 530 535 540 Thr Lys Glu Gly Leu Glu Leu Pro Glu Asp
Glu Glu Glu Lys Lys Lys 545 550 555 560 Gln Glu Glu Lys Lys Thr Lys Phe Glu Asn Leu
Cys Lys Ile Met Lys 565 570 575 Asp Ile Leu Glu Lys Lys Val Glu Lys Val Val Val Ser
Asn Arg Leu 580 585 590 Val Thr Ser Pro Cys Cys Leu Val Thr Ser Thr Tyr Gly Trp Thr
Ala 595 600 605 Asn Met Glu Arg Ile Met Lys Ala Gln Ala Leu Arg Asp Asn Ser Thr 610
615 620 Met Gly Tyr Met Ala Ala Lys Lys His Leu Glu Ile Asn Pro Asp His 625 630 635
640 Ser Ile Ile Glu Thr Leu Arg Gln Lys Ala Glu Ala Asp Lys Asn Asp 645 650 655 Lys
Ser Val Lys Asp Leu Val Ile Leu Leu Tyr Glu Thr Ala Leu Leu 660 665 670 Ser Ser Gly
Phe Ser Leu Glu Asp Pro Gln Thr His Ala Asn Arg Ile 675 680 685 Tyr Arg Met Ile Lys
Leu Gly Leu Gly Ile Asp Glu Asp Asp Pro Thr 690 695 700 Ala Asp Asp Thr Ser Ala Ala
Val Thr Glu Glu Met Pro Leu Glu 705 710 715 720 Gly Asp Asp Asp Thr Ser Arg Met
Glu Glu Val Asp 725 730 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 19 <211>
LENGTH: 1019 <212> TYPE: DNA <213> ORGANISM: Leishmania major <400> SEQUENCE: 19
gaattcggca cgaggtttct gtactttatt gcttccagcc tttattcact cttcgatttc 60 ctctaacc
atgtcctccg agcgcacctt tattgccgtc aagccggacg gctgcagcg 120 cggcctcgtt ggcgagatca
tcgcccgtt cgagcgcaag ggctacaagc tcgtcgctt 180 gaagatactg cagccgacga cggagcaggc
ccagggctac tataaggacc ttgtctcaa 240 gccgttttcc cgggcccttg tgaagtactt ctctctggc
ccgatcgtgt gtatggtgtg 300 ggagggttaag aacgtggtga agagcgccg cgtgctgctc ggcgcgacga
acccggccga 360 ctacagccc ggcacgatcc gtggcgactt tgccgtggat gtgggcccga acgtgtgcc
420 cgggtccgac tctgtggaga gcgcggagcg cgagatcgcc ttttggttca aggcggatga 480
gatcgcgagc tggacgtcg actccgtgtc ccagatctat gagtaacggt gattgcggac 540 acgctttgag
gacgtagctg taccccaat gaattcttct ctgaaaacca catcataagc 600 ctcttaagag gttatttttc
ttgatcgatg cccggtggtg accagcacca ttcctttatc 660 ggattcactc acactcctag cgaatcatgt
agtgcggtga gagtgggctc tggaggagac 720 tgttgtgtag ccatggcttc aggagagaaa acaaaataca
aggaaaaggca atatgtaaact 780 atggggttcc cttttttact atgcaaagtt tttataactc ctgatcggca
aaaacaacaa 840 caaccgccat acaccaagag caaatgcttt cttctgcgga ctgtgcttct gttttttttt
900 atgaaggagt gactcgcgcg atgaaaagtg tgtgcgtggg agatgtattt cttttttttg 960
ttcatagtgg cgacagctca ctggtgacga tgacaaaaaa aaaaaaaaaa aaactcgag 1019 <200>
SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 20 <211> LENGTH: 151 <212> TYPE: PRT
<213> ORGANISM: Leishmania major <400> SEQUENCE: 20 Met Ser Ser Glu Arg Thr Phe Ile
Ala Val Lys Pro Asp Gly Val Gln 1 5 10 15 Arg Gly Leu Val Gly Glu Ile Ile Ala Arg
Phe Glu Arg Lys Gly Tyr 20 25 30 Lys Leu Val Ala Leu Lys Ile Leu Gln Pro Thr Thr
Glu Gln Ala Gln 35 40 45 Gly His Tyr Lys Asp Leu Cys Ser Lys Pro Phe Phe Pro Ala
Leu Val 50 55 60 Lys Tyr Phe Ser Ser Gly Pro Ile Val Cys Met Val Trp Glu Gly Lys 65
70 75 80 Asn Val Val Lys Ser Gly Arg Val Leu Leu Gly Ala Thr Asn Pro Ala 85 90 95
Asp Ser Gln Pro Gly Thr Ile Arg Gly Asp Phe Ala Val Asp Val Gly 100 105 110 Arg Asn
Val Cys His Gly Ser Asp Ser Val Glu Ser Ala Glu Arg Glu 115 120 125 Ile Ala Phe Trp
Phe Lys Ala Asp Glu Ile Ala Ser Trp Thr Ser His 130 135 140 Ser Val Ser Gln Ile Tyr
Glu 145 150 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 21 <211> LENGTH: 1523
<212> TYPE: DNA <213> ORGANISM: Leishmania major <400> SEQUENCE: 21 gaattcggca
cgagtgcgtc ccgacatgac atgctcgtc accggacttc agtgacaga 60 cccgaactgc aagacctgca
caacttacgg tcagtgcaca gactgcaacg acggctacgg 120 tctcacctcc tccagcgttt gctgcgctg
cagtgtagcg ggctgcaaga gctgccccgt 180 cgacgctaac gtctgcaaag tgtgtctcgg cggcagcgag
ccgatcaaca atatgtgcc 240 ctgcaccgac ccaactgcg ccagctgccc cagcgacgct ggcacgtgca
ctcagtgcgc 300 gaacggctac ggtctcgtgg acggcgctg tgtgagatgc caggagccca actgcttcag
360 ctgcgacagc gacgcgaata agtgacaca atgtgcgccc aactactacc taccctcgct 420
cttgacctgc tccccggtgg cctgcaacat cgagcactgc atgcagtgcg acccacagac 480 gccgtcgcgc
tgccaggagt gctgtctccc ctacgtggtt gacagctacg accgctctg 540 caggctctcc gatgctgct
ccgtgcccga ctgcaagaag tgcgagaccg gtacctccag 600 gctctgcgcc gactgcgaca ccggctacag
tctctccgcc gacgcgacga gctgcagcag 660 tccaaccacg cagcgtgcg aggtggagca ctgcaacaca
tgtgtgaacg gcgatagcac 720 ccgctgtgcc tactgcaaca ccggctacta cgtctccgat ggcaagtga
aggccatgca 780 gggctgtac gtgtcgaaact gcgcgcagtg catgctgctt gacagcacca agtgctccac
840 gtgcgtgaaa gggtaacctg tcaagtcgtc ctacagttgc gtctcgcaga aagtcacaa 900
cagtgcggcc gcgcctact ctctgtgggt ggccgcccgc gtgctctca cctcttttgc 960 catgcacct

gcatagtgcg cagcggcatg cgaacaaccc cactctcatt ctccaacatg 1020 tgcatacaca cacacacaga
cagcggggga gcacccctc cccacacaca cacacgcact 1080 tcccccttgt cttgttcttc tttcctcggt
cgcatcttct tctctcgtgc gctggcgccg 1140 gcctcctgca cgtcgctccc ctccccctaa cctctattct
ctctctctct ctctctcgcc 1200 ggcatcattg cttcttacc ctttctgata cttgctcgcg tggcgcgaca
ctgccacagt 1260 cccacagcgc agacacacgt gtttaaacgg cgcaggcatc cctccctatc acttcatttc
1320 tcctaaagcc actcaccaag tcgcacaccg cctccccca tcggcgccgc ttccggggcg 1380
agctgtgagg aatgggtgtg tgctcgacct cgttctcggc agctcactcg catgtgtaca 1440 gccactccaa
ccacgaaagc tctcttctgc gcacataaaa aaaaaaaaaa aaaaaaaact 1500 cgaggggggg cccggtacc
aaa 1523 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 22 <211> LENGTH: 320 <212>
TYPE: PRT <213> ORGANISM: Leishmania major <400> SEQUENCE: 22 Val Leu Pro Asp Met
Thr Cys Ser Leu Thr Gly Leu Gln Cys Thr Asp 1 5 10 15 Pro Asn Cys Lys Thr Cys Thr
Thr Tyr Gly Gln Cys Thr Asp Cys Asn 20 25 30 Asp Gly Tyr Gly Leu Thr Ser Ser Ser
Val Cys Val Arg Cys Ser Val 35 40 45 Ala Gly Cys Lys Ser Cys Pro Val Asp Ala Asn
Val Cys Lys Val Cys 50 55 60 Leu Gly Gly Ser Glu Pro Ile Asn Asn Met Cys Pro Cys
Thr Asp Pro 65 70 75 80 Asn Cys Ala Ser Cys Pro Ser Asp Ala Gly Thr Cys Thr Gln Cys
Ala 85 90 95 Asn Gly Tyr Gly Leu Val Asp Gly Ala Cys Val Arg Cys Gln Glu Pro 100
105 110 Asn Cys Phe Ser Cys Asp Ser Asp Ala Asn Lys Cys Thr Gln Cys Ala 115 120 125
Pro Asn Tyr Tyr Leu Thr Pro Leu Leu Thr Cys Ser Pro Val Ala Cys 130 135 140 Asn Ile
Glu His Cys Met Gln Cys Asp Pro Gln Thr Pro Ser Arg Cys 145 150 155 160 Gln Glu Cys
Val Ser Pro Tyr Val Val Asp Ser Tyr Asp Gly Leu Cys 165 170 175 Arg Leu Ser Asp Ala
Cys Ser Val Pro Asn Cys Lys Lys Cys Glu Thr 180 185 190 Gly Thr Ser Arg Leu Cys Ala
Glu Cys Asp Thr Gly Tyr Ser Leu Ser 195 200 205 Ala Asp Ala Thr Ser Cys Ser Ser Pro
Thr Thr Gln Pro Cys Glu Val 210 215 220 Glu His Cys Asn Thr Cys Val Asn Gly Asp Ser
Thr Arg Cys Ala Tyr 225 230 235 240 Cys Asn Thr Gly Tyr Tyr Val Ser Asp Gly Lys Cys
Lys Ala Met Gln 245 250 255 Gly Cys Tyr Val Ser Asn Cys Ala Gln Cys Met Leu Leu Asp
Ser Thr 260 265 270 Lys Cys Ser Thr Cys Val Lys Gly Tyr Leu Leu Thr Ser Ser Tyr Ser
275 280 285 Cys Val Ser Gln Lys Val Ile Asn Ser Ala Ala Ala Pro Tyr Ser Leu 290 295
300 Trp Val Ala Ala Ala Val Leu Leu Thr Ser Phe Ala Met His Leu Ala 305 310 315 320
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 23 <211> LENGTH: 797 <212> TYPE:
DNA <213> ORGANISM: Leishmania major <400> SEQUENCE: 23 ctgtacttta ttgccaccag
ccagccatgt cctgcggtaa cgccaagatc aactctccc 60 cgccgtcctt cgaggagtg gcgctcatgc
ccaacggcag cttcaagaag atcagcctct 120 cctcctacaa gggcaagtgg gtcgtgctct tcttctacc
gctcgacttt agcttcgtgt 180 gcccgacaga ggtcatcgcg ttctccgaca gcgtgagtcg cttcaacgag
ctcaactgcg 240 aggtcctcgc gtgctcgata gacagcgagt acgcgcacct gcagtggacg ctgcaggacc
300 gcaagaagg cggtcctcgg accatggcga tcccaatgct agccgacaag accaagagca 360
tcgctcgttc ctacggcgtg ctggaggaga gccagggcgt ggctaccgc ggtctcttca 420 tcatcgaccc
ccatggcatg ctgcgtcaga tcaccgtcaa tgacatgccg gtgggccgca 480 gcgtggagga ggttctacgc
ctgctggagg ctttctcagt cgtggagaag cacggcgagg 540 tgtgccccgc gaactggaag aagggcgccc
ccacgatgaa gccggaaccg aatgcgtctg 600 tcgagggata cttcagcaag cagtaaacct gtgagcgtcg
caggatcag tgtgacctca 660 cccgcctctg ccagtgggtg cgagagggcg tgagggattg tgggaaggct
gttgatatg 720 atgcagacag cgatgaatgc aactccaca cactggccct cctcagccct ctccacacag
780 acacacgcac gcattgtg 797 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 24 <211>
LENGTH: 199 <212> TYPE: PRT

Detailed Description Paragraph Table (7):

<213> ORGANISM: Leishmania major <400> SEQUENCE: 24 Met Ser Cys Gly Asn Ala Lys Ile
Asn Ser Pro Ala Pro Ser Phe Glu 1 5 10 15 Glu Val Ala Leu Met Pro Asn Gly Ser Phe
Lys Lys Ile Ser Leu Ser 20 25 30 Ser Tyr Lys Gly Lys Trp Val Val Leu Phe Phe Tyr
Pro Leu Asp Phe 35 40 45 Ser Phe Val Cys Pro Thr Glu Val Ile Ala Phe Ser Asp Ser
Val Ser 50 55 60 Arg Phe Asn Glu Leu Asn Cys Glu Val Leu Ala Cys Ser Ile Asp Ser 65
70 75 80 Glu Tyr Ala His Leu Gln Trp Thr Leu Gln Asp Arg Lys Lys Gly Gly 85 90 95
Leu Gly Thr Met Ala Ile Pro Met Leu Ala Asp Lys Thr Lys Ser Ile 100 105 110 Ala Arg
Ser Tyr Gly Val Leu Glu Glu Ser Gln Gly Val Ala Tyr Arg 115 120 125 Gly Leu Phe Ile
Ile Asp Pro His Gly Met Leu Arg Gln Ile Thr Val 130 135 140 Asn Asp Met Pro Val Gly
Arg Ser Val Glu Glu Val Leu Arg Leu Leu 145 150 155 160 Glu Ala Phe Gln Phe Val Glu
Lys His Gly Glu Val Cys Pro Ala Asn 165 170 175 Trp Lys Lys Gly Ala Pro Thr Met Lys
Pro Glu Pro Asn Ala Ser Val 180 185 190 Glu Gly Tyr Phe Ser Lys Gln 195 <200>
SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 25 <211> LENGTH: 637 <212> TYPE: DNA
<213> ORGANISM: Leishmania tropica <400> SEQUENCE: 25 ttacatatgc atcaccacca
ccaccacatg tctgtcggta acgccaagat caactctccc 60 gcgccgcctt tcgaggagat ggcgctcatg
cccaacggca gcttcaagaa gatcagcctc 120 tccgcctaca agggcaagtg ggtcgtgctc ttcttctacc
cgctcgactt caccttcgtg 180 tgcccgacag agatcatcgc gttctccgac aacgtgagtc gtttcaacga
gctcaactgc 240 gaggtcctcg cgtgctcgat ggacagcgag tacgcgcacc tgcagtggac gctgcaggac
300 cgcaagaagg gcggcctcgg ggccatggcg atcccaatgc tggccgacaa gactaagagc 360

atcgcctcgtt cctacggcgt gctggaggag agccagggcg tggcctaccg cggctctcttc 420 atcatcgacc
cccgtggcat ggtgcgtcag atcaccgtca acgacatgcc ggtggggccgc 480 aacgtggagg aggctctgcg
cctgctggag gctttgcagt tcgtggagaa gcacggcgag 540 gtgtgccccg cgaactggaa gaagggcgcc
cccacgatga agccggaacc gaaggcgtct 600 gtcgaggat acttcagaa gcagtaagaa ttccatg 637
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 26 <211> LENGTH: 206 <212> TYPE:
PRT <213> ORGANISM: Leishmania tropica <400> SEQUENCE: 26 Met His His His His His
His Met Ser Cys Gly Asn Ala Lys Ile Asn 1 5 10 15 Ser Pro Ala Pro Pro Phe Glu Glu
Met Ala Leu Met Pro Asn Gly Ser 20 25 30 Phe Lys Lys Ile Ser Leu Ser Ala Tyr Lys
Gly Lys Trp Val Val Leu 35 40 45 Phe Phe Tyr Pro Leu Asp Phe Thr Phe Val Cys Pro
Thr Glu Ile Ile 50 55 60 Ala Phe Ser Asp Asn Val Ser Arg Phe Asn Glu Leu Asn Cys
Glu Val 65 70 75 80 Leu Ala Cys Ser Met Asp Ser Glu Tyr Ala His Leu Gln Trp Thr Leu
85 90 95 Gln Asp Arg Lys Lys Gly Gly Leu Gly Ala Met Ala Ile Pro Met Leu 100 105
110 Ala Asp Lys Thr Lys Ser Ile Ala Arg Ser Tyr Gly Val Leu Glu Glu 115 120 125 Ser
Gln Gly Val Ala Tyr Arg Gly Leu Phe Ile Ile Asp Pro Arg Gly 130 135 140 Met Val Arg
Gln Ile Thr Val Asn Asp Met Pro Val Gly Arg Asn Val 145 150 155 160 Glu Glu Ala Leu
Arg Leu Leu Glu Ala Leu Gln Phe Val Glu Lys His 165 170 175 Gly Glu Val Cys Pro Ala
Asn Trp Lys Lys Gly Ala Pro Thr Met Lys 180 185 190 Pro Glu Pro Lys Ala Ser Val Glu
Gly Tyr Phe Ser Lys Gln 195 200 205 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO
27 <211> LENGTH: 51 <212> TYPE: DNA <213> ORGANISM: Artificial Sequence <220>
FEATURE: <223> OTHER INFORMATION: PCR primer <400> SEQUENCE: 27 caattacata
tgcatcacca tcaccatcac atgtcctgcg gtaacgccaa g 51 <200> SEQUENCE CHARACTERISTICS:
<210> SEQ ID NO 28 <211> LENGTH: 31 <212> TYPE: DNA <213> ORGANISM: Artificial
Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR primer <400> SEQUENCE: 28
catggaattc ttactgcttg ctgaagtatc c 31 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID
NO 29 <211> LENGTH: 520 <212> TYPE: DNA <213> ORGANISM: Leishmania major <220>
FEATURE: <221> NAME/KEY: misc_feature <222> LOCATION: (1)...(520) <223> OTHER
INFORMATION: n = A, T, C or G <400> SEQUENCE: 29 ggcacgagcc ctgacctaca tttgctcgcc
gatattcgcg gggagttctt caatttgcgt 60 cgcgtagaac tgctcaatgt cgcgcaacaa ggcgagctcg
tcgtggcgca cgaaggtgat 120 ggccagtcca gtgcggccca tgccggccagt gcggccgatg cgggtgaatgt
actgctcacg 180 cgcgagcggc aaatcgtagc tgaggacgag cgagacgcgc tccacatcaa tgccacgcgc
240 ccacaggtcc gttgtaatga ncacgcggct gtgtccatta cggaatgccg cataatctcg 300
tcgcgctccg cctggggcat gtcgccgtgc atggcggaca cagcgaaatt ctgcgcgctc 360 atcttcttgg
caagctgctc cacctttttg cgggtgttgc anaaaaccac ngcgtggcg 420 atcgtaagc tgctgtacaa
actccatcaa gaaatcgaat ttgtttttct cttcgtcnac 480 nganacaaan tactgtttaa cgctntccac
ggtgatctca 520 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 30 <211> LENGTH: 600
<212> TYPE: DNA <213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY:
misc_feature <222> LOCATION: (1)...(600) <223> OTHER INFORMATION: n = A, T, C or G
<400> SEQUENCE: 30 ggcacaaggt tttcgggtta tcttcacgca tgggtggagcg cagatgggtg
aagtaaatac 60 gcggaccgaa ctgcttgatc atatcaacca gatcgttgctc agcacgcacg ccgtangaac 120
cgggtgcacat ggtaaaaccg tntgccatgc tgtttacggt atcaaccatc cactgcatat 180 cttcaatggt
ggaacaatg cgcggcaggc cgaggatccg gcgcggctca tcatnnagnt 240 natnaaccan tcgcacgtct
anttctgcac taaactacaa ntatcggtna catatntaa 300 ggccnatttt cgtccaggaa ntatgtnctn
tcaaaatgcc ncgttannca ctcttaaatg 360 tctcangnngn aaantngttc taaagggtgt ccaaaanntn
nttacnntc ccncttact 420 tcaanantc ctcaattcc cnggcccttn gacnannatt tncattataa
anatanaann 480 ttcaaattna ttcccnacct ncnntnncca aanntancna ataatcann cccntncann
540 anntcccanc ttaccctccn ntngnnggg nnnccnattn ccccaanccc ncnctaaata 600 <200>
SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 31 <211> LENGTH: 600 <212> TYPE: DNA
<213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222>
LOCATION: (1)...(600) <223> OTHER INFORMATION: n = A, T, C or G <400> SEQUENCE: 31
ggcacgagcc tcagtggagc tcaatgaaga tattgcagta tcttactctg gatggcactc 60 aggtctccgg
cacgtgccc cccagtgga gcgcgatggc atcgtgcca attcttaacc 120 tgnagggtac tgaggtctct
ggtacgtgc gcctgaagt gatatcnatg ancaggctgc 180 aaactctgaa tctgcggcg acgaaantat
ccggcactct gccgcccga tgganttcta 240 tgaacagcct ggagtacttt cacccttatc ttactcaggt
ctccggcacg ctgccgccc 300 agtggagtgg gatgtcnaag gccgcatact tctggctgga atactgcgac
ctgtccggca 360 ntctgccgcc cnagtggctg tcnatgcaa agctgcgcgg tatctcactg ancggcaaca
420 aattcttgcg nggtntncc ngactcntgg gattcagaaa ggtggctcct gttgttgggc 480
atcnaaggan caaaccccaa ngggccnncn aattgcttgg gcntgcttaa gganttgac 540 naaccaacnc
cnccaaaaac cccccccacc ncnaaannac nanccccac ttaannccn 600 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 32 <211> LENGTH: 600 <212> TYPE: DNA <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222>
LOCATION: (1)...(600) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 32
ngcacgagaa gcgcaactgg cgcacgcgat ctgtgactat ctgcctgaac aggggcaatn 60 gtttgttgg
aacagcctgg tggtagctt gattgatncg cttnccgaan ttccggcagg 120 ttaccgggtg tacancaacc
gtggggccan cggtatcnac nggtgcttt cgaccgccgc 180 cgnggttcan cgggcaancg gcaaaccgac

gctggcgatt gtgggcgcatc tctccgcact 240 ttacgatctc aacgcncctgg cgttattgcg tcaggtttct
gcgcgcgtgg tattaattgt 300 ggtgaacaac aacggcnggg caaaattttc tcgctgttgc caacgcccc
aaagcnagcg 360 tgaagcggtt ctatctgatg ccgcaaaacg tccattttga aacacgcccgc cncctatgtt
420 tcganctgaa aatatcatcg tccgcaaaac tggcangaaa cttingaaaac cgcattttgc 480
cgacnccctg gcnacgccc aaccaccca ccggttgatt gaaaatgggtg ggtaaacgaa 540 ncnnatggg
tgccccaaan cncnccanc caaattttctg ggcccaggtt aaancccttt 600 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 33 <211> LENGTH: 600 <212> TYPE: DNA <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222>
LOCATION: (1)...(600) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 33
acgatgacca tgccccgaag gaggatggcc atgcgccgaa gaacgatgac catgccccga 60 aggaggatgg
ccatgcgccg aagaacgatg accatgcccc gaaggaggat ggccatgcgc 120 cgaagaacga cggggatgtg
cagaanaaga gcgaagatgg agacaacgtg ggagaggag 180 gcaagggcaa tgaggatggt aacgatgatc
agccgaagga gcacgctgcc ggcaactagt 240 gggctgcgtc cgggcttgtg tgcganccgt gctctgcacc
ccgcgcgtcg tgcattcctc 300 catgtggact gcgtgtgtct ctcccgttt gtctctctcc cccacacagt
ggctgatgcc 360 tgcacggggt tgctgtggct gcacctcctg accactgcc a gtttcttgg cttgcctccc
420 ctctgcgcct ccgctcgtgc cgctcgtgcc gaattcgata tcaagcttat cgataccgtc 480
nacctcgaag gggggccccg ttaccattc gccctatant gactentatt acaattcctg 540 gcgtcgtttt
acacgtcgtg actgggaaaa accctggcgt tccccactta tcgccttgca 600 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 34 <211> LENGTH: 516 <212> TYPE: DNA <213>
ORGANISM: Leishmania major <400> SEQUENCE: 34 <200> SEQUENCE CHARACTERISTICS: <210>
SEQ ID NO 35 <211> LENGTH: 822 <212> TYPE: DNA <213> ORGANISM: Leishmania major
<220> FEATURE: <221> NAME/KEY: misc_feature <222> LOCATION: (1)...(822) <223> OTHER
INFORMATION: n = A,T,C or G <400> SEQUENCE: 35 ggcacganag atcttcgtga agacgtgac
cggcaanacg atcgcgctgg aggtggagcc 60 gagcgacacg atcgagaacg tgaaggcga gatccaggac
aaggagggca tcccgcggga 120 ccagcagcgc ctgatcttcg ccggcaagca gctggaggan ggccgcacgc
tctcggacta 180 caacatccag aaggagtcca cgctgcacct ggtgctgcgc ctgcgcggcg gcatgcanat
240 ctctgtgaaa acgctnaccg gcaanacaat cgcgctggaa gtggagccga acgaccnatc 300
gaaaacgtga aggcnnanac ccangacaag gaaggcctcc cgccgganac gcacgcctga 360 tcttcnccg
gcaaccactt gangaagggc ncacgtctc ngactacnac atccanaaaag 420 gattccnccc tgcaccttgt
tgcttgcncc ttgctcgggg ggcatgccna atcttccttn 480 aaaacctcaa ccggcaanaa caatcccccn
cngaagtgg aacccaacca nccattcna 540 aaactttaaa ggccnnnatt ccngaacaan gaagggcttc
ccccccggac cnncaancnc 600 cctgatntt cccccggnn ncantttgga angaagggcc cncctccn
ccgaattncn 660 acntccnnaa anggattccc cccctnccct tgnnttttgc gccnnnnnnc ggcnncntnc
720 cnaaattccg ncnnaaggnc cccantanac nactttccc ntccccccc nnnnttttgc 780
ntaaantttt tncccccnna aanntccnt ttnncanttn an 822 <200> SEQUENCE CHARACTERISTICS:
<210> SEQ ID NO 36 <211> LENGTH: 146 <212> TYPE: PRT <213> ORGANISM: Leishmania
major <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION: (1)...(146) <223>
OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 36 Gly Thr Ser Pro Cys Leu
His Leu Leu Ala Asp Ile Arg Gly Glu Phe 1 5 10 15 Phe Asn Leu Arg Arg Val Glu Leu
Leu Asn Val Ala Gln Gln Ala Gln 20 25 30 Leu Val Val Ala His Glu Gly Asp Gly Gln
Ser Ser Ala Ala His Ala 35 40 45 Ala Ser Ala Ala Val Asn Val Leu Leu Thr
Arg Glu Arg Gln 50 55 60 Ile Val Ala Glu Asp Glu Arg Asp Ala Leu His Ile Asn Ala
Thr Arg 65 70 75 80 Pro Gln Val Arg Cys Asn Xaa His Ala Ala Val Ser Ile Thr Glu Cys
85 90 95 Arg Ile Ile Ser Ser Arg Ser Ala Trp Gly Met Ser Pro Cys Met Ala 100 105
110 Asp Thr Ala Lys Phe Ser Arg Val Ile Phe Leu Ala Ser Cys Ser Thr 115 120 125 Phe
Leu Arg Val Leu Xaa Lys Thr Thr Ala Trp Ala Ile Val Lys Leu

Detailed Description Paragraph Table (8):

130 135 140 Ser Tyr 145 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 37 <211>
LENGTH: 77 <212> TYPE: PRT <213> ORGANISM: Leishmania major <220> FEATURE: <221>
NAME/KEY: VARIANT <222> LOCATION: (1)...(77) <223> OTHER INFORMATION: Xaa = Any
Amino Acid <400> SEQUENCE: 37 Ala Gln Gly Phe Arg Val Ile Phe Thr His Gly Gly Ala
Gln Met Gly 1 5 10 15 Glu Val Asn Thr Arg Thr Glu Leu Leu Asp His Ile Asn Gln Ile
Val 20 25 30 Val Ser Thr His Ala Val Xaa Thr Gly Ala His Gly Lys Thr Val Cys 35 40
45 His Ala Val Tyr Gly Ile Asn His Pro Leu His Ile Phe Asn Gly Gly 50 55 60 Asn Asn
Ala Arg Gln Ala Glu Asp Pro Ala Arg Leu Ile 65 70 75 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 38 <211> LENGTH: 68 <212> TYPE: PRT <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION:
(1)...(68) <223> OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 38 His Glu
Pro Gln Trp Ser Ser Met Lys Ile Leu Gln Tyr Leu Thr Leu 1 5 10 15 Asp Gly Thr Gln
Val Ser Gly Thr Leu Pro Pro Gln Trp Ser Ala Met 20 25 30 Ala Ser Val Arg Ile Leu
Asn Leu Xaa Gly Thr Glu Val Ser Gly Thr 35 40 45 Leu Pro Pro Glu Trp Ile Ser Met
Xaa Arg Leu Gln Thr Leu Asn Leu 50 55 60 Arg Arg Thr Lys 65 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 39 <211> LENGTH: 65 <212> TYPE: PRT <213>

ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION: (1)...(65) <223> OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 39 Ala Arg Glu Ala Gln Leu Ala His Arg Ile Cys Asp Tyr Leu Pro Glu 1 5 10 15 Gln Gly Gln Xaa Phe Val Gly Asn Ser Leu Val Val Arg Leu Ile Asp 20 25 30 Xaa Leu Xaa Gln Xaa Pro Ala Gly Tyr Pro Val Tyr Xaa Asn Arg Gly 35 40 45 Ala Xaa Gly Ile Xaa Xaa Leu Leu Ser Thr Ala Ala Gly Val Xaa Arg 50 55 60 Ala 65 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 40 <211> LENGTH: 78 <212> TYPE: PRT <213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION: (1)...(78) <223> OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 40 Asp Asp His Ala Pro Lys Glu Asp Gly His Ala Pro Lys Asn Asp Asp 1 5 10 15 His Ala Pro Lys Glu Asp Gly His Ala Pro Lys Asn Asp Asp His Ala 20 25 30 Pro Lys Glu Asp Gly His Ala Pro Lys Asn Asp Gly Asp Val Gln Xaa 35 40 45 Lys Ser Glu Asp Gly Asp Asn Val Gly Glu Gly Gly Lys Gly Asn Glu 50 55 60 Asp Gly Asn Asp Asp Gln Pro Lys Glu His Ala Ala Gly Asn 65 70 75 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 41 <211> LENGTH: 169 <212> TYPE: PRT <213> ORGANISM: Leishmania major <400> SEQUENCE: 41 Leu Gln Gln Arg Leu Asp Thr Ala Thr Gln Gln Arg Ala Glu Leu Glu 1 5 10 15 Ala Arg Val Ala Arg Leu Ala Ala Asp Arg Asp Glu Ala Arg Gln Gln 20 25 30 Leu Ala Ala Asn Ala Glu Glu Leu Gln Gln Arg Leu Asp Thr Ala Thr 35 40 45 Gln Gln Arg Ala Glu Leu Glu Ala Arg Val Ala Arg Leu Ala Ala Asp 50 55 60 Gly Asp Glu Ala Arg Gln Gln Leu Ala Ala Asn Ala Glu Glu Leu Gln 65 70 75 80 Gln Arg Leu Asp Thr Ala Thr Gln Gln Arg Ala Glu Leu Glu Ala Gln 85 90 95 Val Ala Arg Leu Ala Ala Asn Ala Glu Glu Leu Gln Gln Arg Leu Asp 100 105 110 Thr Ala Thr Gln Gln Arg Ala Glu Leu Glu Ala Arg Val Ala Arg Leu 115 120 125 Ala Ala Asp Arg Asp Glu Ala Arg Gln Gln Leu Ala Ala Asn Ala Glu 130 135 140 Glu Leu Gln Gln Arg Leu Asp Thr Ala Thr Gln Gln Arg Ala Glu Leu 145 150 155 160 Glu Ala Gln Val Ala Arg Leu Ala Ala 165 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 42 <211> LENGTH: 98 <212> TYPE: PRT <213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION: (1)...(98) <223> OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 42 Ala Arg Xaa Ile Phe Val Lys Thr Leu Thr Gly Xaa Thr Ile Ala Leu 1 5 10 15 Glu Val Glu Pro Ser Asp Thr Ile Glu Asn Val Lys Ala Lys Ile Gln 20 25 30 Asp Lys Glu Gly Ile Pro Pro Asp Gln Gln Arg Leu Ile Phe Ala Gly 35 40 45 Lys Gln Leu Glu Xaa Gly Arg Thr Leu Ser Asp Tyr Asn Ile Gln Lys 50 55 60 Glu Ser Thr Leu His Leu Val Leu Arg Leu Arg Gly Gly Met Xaa Ile 65 70 75 80 Phe Val Lys Thr Leu Thr Gly Xaa Thr Ile Ala Leu Glu Val Glu Pro 85 90 95 Asn Asp <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 43 <211> LENGTH: 39 <212> TYPE: PRT <213> ORGANISM: Leishmania major <400> SEQUENCE: 43 Leu Gln Gln Arg Leu Asp Thr Ala Thr Gln Gln Arg Ala Glu Leu Glu 1 5 10 15 Ala Arg Val Ala Arg Leu Ala Ala Asp Arg Asp Glu Ala Arg Gln Gln 20 25 30 Leu Ala Ala Asn Ala Glu Glu 35 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 44 <211> LENGTH: 600 <212> TYPE: DNA <213> ORGANISM: Leishmania chagasi <220> FEATURE: <221> NAME/KEY: misc feature <222> LOCATION: (1)...(600) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 44 cggccgcctc agcaggagg agatcgagcg catgtgtcgc gaggtgccc agttcgagga 60 tgaggaccgc aaggtgctgc aacgtgtcga agcgaagaac tcgctagaga gcatcgcgta 120 ctgccttcgc aaccagatca acgacaagga caagcttggt gacaagctcg ccgcggagca 180 caagaaggcg atcgaggagg ctgtgaagga tgccctcgac tttgtccacg agaaccctca 240 tgcagaccgt gaggagtctg aggctgctcg cacgaagctg cagagtgtga cgaaccccat 300 cattcaaaag gtgtaccagg gcgcgcgcg ctctggtgca gaagaggcgg acgcgatgga 360 tgacttggtta gtcggccgcg tgaagaagaaa aacagggaaa gcgggaacat nccacaanaa 420 ccnaagaaga aagggggtng cgacaccgct cgaacaccga cggcncacat ncntcatggg 480 catgctcagc tttcctctcc ccaacaaacc agaaggtttt ctccaaacnc cgtctcngcn 540 cccaaaatac ggaaangtta ancgaaaaan ccccttccac caattgnngt tcttttgttt 600 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 45 <211> LENGTH: 1748 <212> TYPE: DNA <213> ORGANISM: Leishmania chagasi <400> SEQUENCE: 45 ctagtggatc ccccgggctg caggaattca cggaatacgt acctcctccc ctttcttggt 60 agaagaacaa caacaacgtt caagacgacg ccgcgccttc ttgtaccgca tttgcttctg 120 agcacgttca atccgtgcct tgcaaacatg gaggcgtaca agaagctgga aacgatcttt 180 acgaaggtct accgcctgga ccatttctc ggtctgggca actgggacat gaacacaaac 240 atgcccccca agggcgagga atcacgcggt gaggcgatgg cgatgctctc ggagctccgc 300 tttggcttca tcacggcacc ggaggtgaaa agcctgattg agagtgccac caagggcagc 360 gaggagctga atgcggtgca gcgcgctaac ttgcgggaga tgaggcgtgc gtggaagagc 420 gccaccgcct tgccggctga gtttgtgggc cgcaagatgc gcctcacgac acacgcgcac 480 agcgtgtggc gcgacagccg caaagcaaat gacttcgcca agttcttacc ggtgctcagg 540 gacctggttg cgctcgccc tgaggagggc tcatacctcg ccgcccggcac ctccctctcc 600 ccgtatgagg cgctcatgaa cgagtacgag ccaggaaatca cgacacaaaa gctggatgag 660 gtgtacgcaa atgtaaagtc gtggctgccg gacgtgctaa aggacattgt gcagaagcag 720 tccggcgagt cggtgattgc gttctcgcat aagttccgc aggacaagca ggaagcactg 780 tgcaaggaat tcatgaagat ctggcacttc gacaccgatg ccggtgcct cgacgtcagc 840 cccaccctt tcacgggaat gacgaaggag gactgccgac tcacaacaaa ctacatcgaa 900 gacacgtttg ttcagagctt

Detailed Description Paragraph Table (9):

h e b b g e e e f c e e g e

Lys Val Tyr Gln Gly Ala Ala Gly Ser Gly 100 105 110 Ala Glu Glu Ala Asp Ala Met Asp
Asp Leu Leu Val Gly Arg Val Lys 115 120 125 Arg Lys Thr Gly Lys Ala Gly Thr 130 135
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 50 <211> LENGTH: 510 <212> TYPE:
PRT <213> ORGANISM: Leishmania chagasi <400> SEQUENCE: 50 Tyr Leu Leu Pro Leu Leu
Gly Arg Arg Thr Thr Thr Thr Phe Lys Thr 1 5 10 15 Thr Pro Arg Leu Leu Val Pro His
Leu Leu Leu Ser Thr Phe Asn Pro 20 25 30 Cys Leu Ala Asn Met Glu Ala Tyr Lys Lys
Leu Glu Thr Ile Phe Thr 35 40 45 Lys Val Tyr Arg Leu Asp His Phe Leu Gly Leu Gly
Asn Trp Asp Met 50 55 60 Asn Thr Asn Met Pro Pro Lys Gly Glu Glu Ser Arg Gly Glu
Ala Met 65 70 75 80 Ala Met Leu Ser Glu Leu Arg Phe Gly Phe Ile Thr Ala Pro Glu Val
85 90 95 Lys Ser Leu Ile Glu Ser Ala Thr Lys Gly Ser Glu Glu Leu Asn Ala 100 105
110 Val Gln Arg Ala Asn Leu Arg Glu Met Arg Arg Ala Trp Lys Ser Ala 115 120 125 Thr
Ala Leu Pro Ala Glu Phe Val Gly Arg Lys Met Arg Leu Thr Thr 130 135 140 His Ala His
Ser Val Trp Arg Asp Ser Arg Lys Ala Asn Asp Phe Ala 145 150 155 160 Lys Phe Leu Pro
Val Leu Arg Asp Leu Val Ala Leu Ala Arg Glu Glu 165 170 175 Gly Ser Tyr Leu Ala Ala
Gly Thr Ser Leu Ser Pro Tyr Glu Ala Leu 180 185 190 Met Asn Glu Tyr Glu Pro Gly Ile
Thr Thr Gln Lys Leu Asp Glu Val 195 200 205 Tyr Ala Asn Val Lys Ser Trp Leu Pro Gln
Leu Leu Lys Asp Ile Val 210 215 220 Gln Lys Gln Ser Gly Glu Ser Val Ile Ala Phe Ser
His Lys Phe Pro 225 230 235 240 Gln Asp Lys Gln Glu Ala Leu Cys Lys Glu Phe Met Lys
Ile Trp His 245 250 255 Phe Asp Thr Asp Ala Gly Arg Leu Asp Val Ser Pro His Pro Phe
Thr 260 265 270 Gly Met Thr Lys Glu Asp Cys Arg Leu Thr Thr Asn Tyr Ile Glu Asp 275
280 285 Thr Phe Val Gln Ser Leu Tyr Gly Val Ile His Glu Ser Gly His Gly 290 295 300
Lys Tyr Glu Gln Asn Cys Gly Pro Arg Glu His Ile Thr Gln Pro Val 305 310 315 320 Cys
Asn Ala Arg Ser Leu Gly Leu His Glu Ser Gln Ser Leu Phe Ala 325 330 335 Glu Phe Gln
Ile Gly His Ala Thr Pro Phe Ile Asp Tyr Leu Thr Thr 340 345 350 Arg Leu Pro Glu Phe
Phe Glu Ala Gln Pro Ala Phe Ser Gln Asp Asn 355 360 365 Met Arg Lys Ser Leu Gln Gln
Val Lys Pro Gly Tyr Ile Arg Val Asp 370 375 380 Ala Asp Glu Val Cys Tyr Pro Leu His
Val Ile Leu Arg Tyr Glu Ile 385 390 395 400 Glu Arg Asp Leu Met Glu Gly Lys Met Glu
Val Glu Asp Val Pro Arg 405 410 415 Ala Trp Asn Ala Lys Met Gln Glu Tyr Leu Gly Leu
Ser Thr Glu Gly 420 425 430 Arg Asp Asp Val Gly Cys Leu Gln Asp Val His Trp Ser Met
Val Arg 435 440 445 Ser Ala Thr Leu Arg Arg Thr Arg Ser Ala Pro Cys Met Arg Arg Arg
450 455 460 Ser Trp Arg Ala Ser Glu Arg Ser Trp Glu Thr Thr Arg Trp Met Ser 465 470
475 480 Ala Cys Ala Pro Val Ser Ser Ala Pro Ser Trp Lys Ser Ser Arg Arg 485 490 495
Arg Ser Gly Ile Met Gly Ala Cys Thr Arg Arg Thr Thr Ser 500 505 510 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 51 <211> LENGTH: 107 <212> TYPE: PRT <213>
ORGANISM: Leishmania chagasi <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION:
(1)...(107) <223> OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 51 Gly
Arg Arg Met Ala Ile His Arg Lys Met Thr Ala Met Ala Leu Arg 1 5 10 15 Arg Thr Ala
Val His Arg Lys Thr Thr Thr Val Ala Leu Arg Arg Thr 20 25 30 Ala Ile His Arg Lys
Met Thr Ala Met Ala Leu Arg Arg Thr Ala Val 35 40 45 His Arg Lys Ile Thr Ala Met
Ala Leu Arg Arg Thr Ala Ile His Arg 50 55 60 Lys Met Thr Ala Met Pro Leu Arg Arg
Thr Ala Val His Xaa Lys Met 65 70 75 80 Thr Ala Met Ala Leu Arg Arg Thr Ala Val His
Arg Lys Met Thr Ala 85 90 95 Met Ala Leu Arg Xaa Thr Pro Tyr Thr Glu Lys 100 105
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 52 <211> LENGTH: 63 <212> TYPE: PRT
<213> ORGANISM: Leishmania chagasi <400> SEQUENCE: 52 Val Arg Glu Thr Arg Thr Cys
Met Leu Leu Val Glu Gly Ala Glu Leu 1 5 10 15 Val Pro Leu Met His Lys His Glu Val
Val His Cys Leu Val Arg His 20 25 30 Phe Pro Glu His Asn Glu Gln Arg His His Thr
Ala Ser Asn Ser Arg 35 40 45 Gln Ser Glu Cys Asp Ala His Thr Lys Ser Lys Val Val
Asp Gln 50 55 60 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 53 <211> LENGTH:
324 <212> TYPE: PRT <213> ORGANISM: Leishmania chagasi <220> FEATURE: <221>
NAME/KEY: VARIANT <222> LOCATION: (1)...(324) <223> OTHER INFORMATION: Xaa = Any
Amino Acid <400> SEQUENCE: 53 Phe Arg Gly Ile Pro Arg Asn Ser Val Arg Arg Gly Thr
Arg Thr Gly 1 5 10 15 Val Asp Ser Asp Ala Thr Ser Ser Thr Thr Ala Ala Tyr Asp Gly
Ala 20 25 30 Gly Ser Ala Pro Val Met Val Asp Ala Asn Val Ser His Pro Pro Tyr 35 40
45 Ala Gly His Asp Gln Val Tyr Met His Val Gly Lys Pro Ile Val Gly 50 55 60 Asn Thr
Leu Asp Gly Tyr Asn Gly Cys Val Phe Ala Tyr Gly Xaa Thr 65 70 75 80 Gly Ser Gly Lys
Thr Phe Thr Met Leu Gly Tyr Ala Pro Ser Thr Xaa 85 90 95 Asp Ile Arg Ala Arg Lys
Gly Ser Val Pro Cys Gly Ala Ser Ser Met 100 105 110 Glu Asn Ser Thr Pro Leu Asp Ser
Ala Val Glu Pro Phe Glu Ser Asp 115 120 125 Asp Gly Asp Asp Val Val Asp Lys Thr Gly
Leu Asp Pro Asn Glu Leu 130 135 140 Gln Gly Ile Ile Pro Arg Ala Cys Thr Asp Leu Phe
Asp Gly Leu Arg 145 150 155 160 Ala Lys Arg Ala Lys Asp Ser Phe Thr Tyr Arg Val
Glu Val Ser 165 170 175 Tyr Tyr Glu Ile Tyr Asn Glu Lys Val Phe Asp Leu Ile Arg Pro
Gln 180 185 190 Arg Asn Thr Asp Leu Arg Ile Arg Asn Ser Pro Asn Ser Gly Pro Phe 195
200 205 Ile Glu Gly Leu Thr Trp Lys Met Val Ser Lys Glu Glu Asp Val Ala 210 215 220
Arg Val Ile Arg Lys Gly Met Gln Glu Arg His Thr Ala Ala Thr Lys 225 230 235 240 Phe

Asn Asp Arg Ser Ser Arg Ser His Ala Ile Leu Thr Phe Asn Ile 245 250 255 Val Gln Leu
Ser Met Asp Asp Ser Asp Asn Ala Phe Gln Met Arg Ser 260 265 270 Lys Leu Asn Leu Val
Asp Leu Ala Gly Ser Glu Arg Thr Gly Ala Ala 275 280 285 Gly Ala Glu Gly Asn Glu Phe
His Asp Gly Val Lys Ile Asn His Ser 290 295 300 Leu Thr Val Leu Gly Arg Val Ile Asp
Arg Leu Ala Asp Leu Ser Gln 305 310 315 320 Asn Lys Gly Gly <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 54 <211> LENGTH: 1585 <212> TYPE: DNA <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222>
LOCATION: (1)...(1585) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 54
aaagctggag ctccaccgcg gtggcgggcg ctctagaact agtggatccc ccgggctgca 60 ggaattcggc
acgagtgtcg cccgacatga catgctcgct gaccggactt cagtgcacag 120 acccgaactg caagacctgc
acaacttacg gtcagtgcac agactgcaac gacggctacg 180 gtctcacctc ctccagcgtt tgcgtgctgc
gcagtgtagc ggggtgcaag agctgccccg 240 tcgacgctaa cgtctgcaaa gtgtgtctcg gcggcagcga
gccgatcaac aatatgtgcc 300 cctgcaccga ccccaactgc gccagctgcc ccagcgacgc tggcacgtgc
actcagtgcg 360 cgaacggcta cggctctcgtg gacggcgccct gtgtgagatg ccaggagccc aactgcttca
420 gctgcgacag cgacgcgaat aagtgcacac aatgtgcgcc gaactactac ctcaccccg 480
tcttgacctg ctccccggtg gcctgcaaca tcgagcactg catgcagtgc gaccacaga 540 cgccgtcgcg
ctgccaggag tgcgtgtccc cctacgtggt tgacagctac gacggcctct 600 gcaggctctc cgatgcctgc
tccgtgccc actgcaagaa gtgcgagacc ggtacctcca 660 ggctctgcgc cgagtgcgac accggctaca
gtctctccgc cgacgcgacg agctgcagca 720 gtccaaccac gcagccgtgc gaggtggagc actgcaacac
atgtgtgaac ggcgatagca 780 cccgctgtgc ctactgcaac accggctact acgtctccga tggcaagtgc
aaggccatgc 840 agggctgcta cgtgtcgaac tgcgcgcagt gcatgtgct tgacagcacc aagtgtcca
900 cgtgcgtgaa agggctacctg ctcacgtcgt cctacagttg cgtctcgag aaagtcatca 960
acagtgcggc cgccgctac tctctgtggg tggccgcgcg cgtctcctc acctctttg 1020 ccatgcacct
agcatagtgc gcagcggcat gcgaacaacc ccaactctcat tctccaacat 1080 gtgcatacac acacacacag
acagcggggc agcaccacct cccacacac acacacgcac 1140 ttcccccttg tctgttctt ctttcctcgn
ttcgatttc tttctctcgt gcgtgtggcg 1200 cggcctctg cacgtcgctc cctccccct aacctctatt
ctctctctct ctctctctcg 1260 ccggcatcat tgcttcttac cttttctga tccttgctcg cgtgggcgga
cactgccaca 1320 gtcccacagc gcagacacac gtgtttaaac ggcgcaggca tccctcccta tcacttcatt
1380 tctcctaaag ccactcacca agtcgcacac cgccctcccc catcgccgc ccttcggggc 1440
gcagctgtgc ggaatgggtg tgtgtctgac ctcttctctg gcagctcact cgcattgtga 1500 cagccactcc
aaccacgaaa gctctcttct gcgcacataa aaaaaaaaaa aaaaaaaaaa 1560 ctcgaggggg ggcccggtac
ccaaa 1585 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 55 <211> LENGTH: 320
<212> TYPE: PRT <213> ORGANISM: Leishmania major <400> SEQUENCE: 55 Val Leu Pro Asp
Met Thr Cys Ser Leu Thr Gly Leu Gln Cys Thr Asp 1 5 10 15 Pro Asn Cys Lys Thr Cys
Thr Thr Tyr Gly Gln Cys Thr Asp Cys Asn 20 25 30 Asp Gly Tyr Gly Leu Thr Ser Ser
Ser Val Cys Val Arg Cys Ser Val 35 40 45

Detailed Description Paragraph Table (10):

Ala Gly Cys Lys Ser Cys Pro Val Asp Ala Asn Val Cys Lys Val Cys 50 55 60 Leu Gly
Gly Ser Glu Pro Ile Asn Asn Met Cys Pro Cys Thr Asp Pro 65 70 75 80 Asn Cys Ala Ser
Cys Pro Ser Asp Ala Gly Thr Cys Thr Gln Cys Ala 85 90 95 Asn Gly Tyr Gly Leu Val
Asp Gly Ala Cys Val Arg Cys Gln Glu Pro 100 105 110 Asn Cys Phe Ser Cys Asp Ser Asp
Ala Asn Lys Cys Thr Gln Cys Ala 115 120 125 Pro Asn Tyr Tyr Leu Thr Pro Leu Leu Thr
Cys Ser Pro Val Ala Cys 130 135 140 Asn Ile Glu His Cys Met Gln Cys Asp Pro Gln Thr
Pro Ser Arg Cys 145 150 155 160 Gln Glu Cys Val Ser Pro Tyr Val Val Asp Ser Tyr Asp
Gly Leu Cys 165 170 175 Arg Leu Ser Asp Ala Cys Ser Val Pro Asn Cys Lys Lys Cys Glu
Thr 180 185 190 Gly Thr Ser Arg Leu Cys Ala Glu Cys Asp Thr Gly Tyr Ser Leu Ser 195
200 205 Ala Asp Ala Thr Ser Cys Ser Ser Pro Thr Thr Gln Pro Cys Glu Val 210 215 220
Glu His Cys Asn Thr Cys Val Asn Gly Asp Ser Thr Arg Cys Ala Tyr 225 230 235 240 Cys
Asn Thr Gly Tyr Tyr Val Ser Asp Gly Lys Cys Lys Ala Met Gln 245 250 255 Gly Cys Tyr
Val Ser Asn Cys Ala Gln Cys Met Leu Leu Asp Ser Thr 260 265 270 Lys Cys Ser Thr Cys
Val Lys Gly Tyr Leu Leu Thr Ser Ser Tyr Ser 275 280 285 Cys Val Ser Gln Lys Val Ile
Asn Ser Ala Ala Ala Pro Tyr Ser Leu 290 295 300 Trp Val Ala Ala Ala Val Leu Leu Thr
Ser Phe Ala Met His Leu Ala 305 310 315 320 <200> SEQUENCE CHARACTERISTICS: <210>
SEQ ID NO 56 <211> LENGTH: 14 <212> TYPE: PRT <213> ORGANISM: Leishmania major
<400> SEQUENCE: 56 Pro Lys Glu Asp Gly His Ala Pro Lys Asn Asp Asp His Ala 1 5 10
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 57 <211> LENGTH: 7 <212> TYPE: PRT
<213> ORGANISM: Leishmania major <400> SEQUENCE: 57 Pro Lys Glu Asp Gly His Ala 1 5
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 58 <211> LENGTH: 7 <212> TYPE: PRT
<213> ORGANISM: Leishmania major <400> SEQUENCE: 58 Pro Lys Asn Asp His Ala 1 5
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 59 <211> LENGTH: 264 <212> TYPE:
DNA <213> ORGANISM: Leishmania chagasi <400> SEQUENCE: 59 atgcaccatc atcaccatca
catgggaagc tcctgcacga aggactccgc aaaggagccc 60 cagaagcgtg ctgataacat cgatacgacc
actcgaagcg atgagaagga cggcatccat 120 gtccaggaga gcgccgggtcc tgtgcaggag aacttcgggg

atgctgcagga gaagaacgaa 180 gatggacaca acgtggggga tggagctaac gacaatgagg atggtaacga
tgatcagccg 240 aaggagcagg ttgccggcaa ctag 264 <200> SEQUENCE CHARACTERISTICS: <210>
SEQ ID NO 60 <211> LENGTH: 744 <212> TYPE: DNA <213> ORGANISM: Leishmania chagasi
<400> SEQUENCE: 60 atgggagcct actgcacgaa ggactccgca aaggagcccc agaagcgtgc
tgataacatc 60 cataaaacca ctgaggccaa tcacagaggc gccgccgtg tgccccgaa gcacgccggc 120
ggtgcgatga acgactctgc cccgaaggag gatggccata cacagaaaaa tgacggcgat 180 ggcctaagg
aggacggccg tacacagaaa aacgacgacg gtggccctaa ggaggacggc 240 catacacaga aaaatgacgg
cgatggccct aaggaggacg gccgtacaca gaaaaataac 300 ggcgatggcc ctaaggagga cggccataca
cagaaaaatg acggcgatgc ccctaaggag 360 gacggccgta cacagaaaaa tgacggcgat ggcctaagg
aggacggccg tacacagaaa 420 aatgacggcg atggccctaa ggaggacggc cgtaacacaga aaaatgacgg
cgatggccct 480 aaggaggacg gccgtacaca gaaaaatgac ggcgatggcc ctaaggagga cggccataca
540 cagaaaaatg acggcgatgg ccctaaggag gacggccgta cacagaaaaa tgacggcggt 600
ggcctaagg aggatgagaa tctgcagcaa aacgatggga atgctgcagga gaagaacgaa 660 gatggacaca
acgtggggga tggagctaac ggaatgagg atggtaacga tgatcagccg 720 aaggagcagg ttgccggcaa
ctag 744 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 61 <211> LENGTH: 80 <212>
TYPE: PRT <213> ORGANISM: Leishmania chagasi <400> SEQUENCE: 61 Met Gly Ser Ser Cys
Thr Lys Asp Ser Ala Lys Glu Pro Gln Lys Arg 1 5 10 15 Ala Asp Asn Ile Asp Thr Thr
Thr Arg Ser Asp Glu Lys Asp Gly Ile 20 25 30 His Val Gln Glu Ser Ala Gly Pro Val
Gln Glu Asn Phe Gly Asp Ala 35 40 45 Gln Glu Lys Asn Glu Asp Gly His Asn Val Gly
Asp Gly Ala Asn Asp 50 55 60 Asn Glu Asp Gly Asn Asp Asp Gln Pro Lys Glu Gln Val
Ala Gly Asn 65 70 75 80 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 62 <211>
LENGTH: 247 <212> TYPE: PRT <213> ORGANISM: Leishmania chagasi <400> SEQUENCE: 62
Met Gly Ala Tyr Cys Thr Lys Asp Ser Ala Lys Glu Pro Gln Lys Arg 1 5 10 15 Ala Asp
Asn Ile His Lys Thr Thr Glu Ala Asn His Arg Gly Ala Ala 20 25 30 Gly Val Pro Pro
Lys His Ala Gly Gly Ala Met Asn Asp Ser Ala Pro 35 40 45 Lys Glu Asp Gly His Thr
Gln Lys Asn Asp Gly Asp Gly Pro Lys Glu 50 55 60 Asp Gly Arg Thr Gln Lys Asn Asp
Asp Gly Gly Pro Lys Glu Asp Gly 65 70 75 80 His Thr Gln Lys Asn Asp Gly Asp Gly Pro
Lys Glu Asp Gly Arg Thr 85 90 95 Gln Lys Asn Asn Gly Asp Gly Pro Lys Glu Asp Gly
His Thr Gln Lys 100 105 110 Asn Asp Gly Asp Ala Pro Lys Glu Asp Gly Arg Thr Gln Lys
Asn Asp 115 120 125 Gly Asp Gly Pro Lys Glu Asp Gly Arg Thr Gln Lys Asn Asp Gly Asp
130 135 140 Gly Pro Lys Glu Asp Gly Arg Thr Gln Lys Asn Asp Gly Asp Gly Pro 145 150
155 160 Lys Glu Asp Gly Arg Thr Gln Lys Asn Asp Gly Asp Gly Pro Lys Glu 165 170 175
Asp Gly His Thr Gln Lys Asn Asp Gly Asp Gly Pro Lys Glu Asp Gly 180 185 190 Arg Thr
Gln Lys Asn Asp Gly Gly Gly Pro Lys Glu Asp Glu Asn Leu 195 200 205 Gln Gln Asn Asp
Gly Asn Ala Gln Glu Lys Asn Glu Asp Gly His Asn 210 215 220 Val Gly Asp Gly Ala Asn
Gly Asn Glu Asp Gly Asn Asp Asp Gln Pro 225 230 235 240 Lys Glu Gln Val Ala Gly Asn
245 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 63 <211> LENGTH: 14 <212> TYPE:
PRT <213> ORGANISM: Leishmania chagasi <220> FEATURE: <221> NAME/KEY: VARIANT <222>
LOCATION: (6)...(6) <223> OTHER INFORMATION: Xaa = His or Arg <400> SEQUENCE: 63
Pro Lys Glu Asp Gly Xaa Thr Gln Lys Asn Asp Xaa Gly 1 5 10 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 64 <211> LENGTH: 7 <212> TYPE: PRT <213> ORGANISM:
Leishmania chagasi <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION: (6)...(6)
<223> OTHER INFORMATION: Xaa = His or Arg <400> SEQUENCE: 64 Pro Lys Glu Asp Gly
Xaa Thr 1 5 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 65 <211> LENGTH: 7
<212> TYPE: PRT <213> ORGANISM: Leishmania chagasi <220> FEATURE: <221> NAME/KEY:
VARIANT <222> LOCATION: (5)...(5) <223> OTHER INFORMATION: Xaa = Gly or Asp <400>
SEQUENCE: 65 Gln Lys Asn Asp Xaa Xaa Gly 1 5 <200> SEQUENCE CHARACTERISTICS: <210>
SEQ ID NO 66 <211> LENGTH: 17 <212> TYPE: PRT <213> ORGANISM: Artificial Sequence
<220> FEATURE: <223> OTHER INFORMATION: Synthetic peptide to asses diagnostic
potential of repeat in Lc Gene B <400> SEQUENCE: 66 Gly Cys Gly Pro Lys Glu Asp Gly
Arg Thr Gln Lys Asn Asp Gly Asp 1 5 10 15 Gly <200> SEQUENCE CHARACTERISTICS: <210>
SEQ ID NO 67 <211> LENGTH: 31 <212> TYPE: PRT <213> ORGANISM: Artificial Sequence
<220> FEATURE: <223> OTHER INFORMATION: Synthetic peptide to asses diagnostic
potential of repeat in Lc Gene B <400> SEQUENCE: 67 Gly Cys Gly Pro Lys Glu Asp Gly
Arg Thr Gln Lys Asn Asp Gly Asp 1 5 10 15 Gly Pro Lys Glu Asp Gly Arg Thr Gln Lys
Asn Asp Gly Asp Gly 20 25 30 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 68
<211> LENGTH: 45 <212> TYPE: PRT <213> ORGANISM: Artificial Sequence <220> FEATURE:
<223> OTHER INFORMATION: Synthetic peptide to asses diagnostic potential of repeat
in Lc Gene B <400> SEQUENCE: 68 Gly Cys Gly Pro Lys Glu Asp Gly Arg Thr Gln Lys Asn
Asp Gly Asp 1 5 10 15 Gly Pro Lys Glu Asp Gly Arg Thr Gln Lys Asn Asp Gly Asp Gly
Pro 20 25 30 Lys Glu Asp Gly Arg Thr Gln Lys Asn Asp Gly Asp Gly 35 40 45 <200>
SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 69 <211> LENGTH: 17 <212> TYPE: PRT <213>
ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: Synthetic
peptide to asses diagnostic potential of repeat in Lc Gene B <400> SEQUENCE: 69 Gly

Cys Gly Pro Lys Glu Asp Gly His Thr Gln Lys Asn Asp Gly Asp 1 5 10 15 Gly <200>
SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 70 <211> LENGTH: 31 <212> TYPE: PRT <213>
ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: Synthetic
peptide to asses diagnostic potential of repeat in Lc Gene B <400> SEQUENCE: 70 Gly
Cys Gly Pro Lys Glu Asp Gly His Thr Gln Lys Asn Asp Gly Asp 1 5 10 15 Gly Pro Lys
Glu Asp Gly His Thr Gln Lys Asn Asp Gly Asp Gly 20 25 30 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 71 <211> LENGTH: 45

Detailed Description Paragraph Table (11):

<212> TYPE: PRT <213> ORGANISM: Gly Cys Gly Pro Lys Glu Asp Gly His Thr Gln <220>
FEATURE: <223> OTHER INFORMATION: Synthetic peptide to asses diagnostic potential
of repeat in Lc Gene B <400> SEQUENCE: 71 Gly Cys Gly Pro Lys Glu Asp Gly His Thr
Gln Lys Asn Asp Gly Asp 1 5 10 15 Gly Pro Lys Glu Asp Gly His Thr Gln Lys Asn Asp
Gly Asp Gly Pro 20 25 30 Lys Glu Asp Gly His Thr Gln Lys Asn Asp Gly Asp Gly 35 40
45 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 72 <211> LENGTH: 664 <212> TYPE:
DNA <213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature
<222> LOCATION: (1)...(664) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE:
72 gctgcaggaa ttcggcacga gattgcttcc cagcccacct tcgctatcca gccactctcg 60 ctcttctaca
tctcccaccc cctcacaccg ccatggcttc ttcccgaag gcttccaacc 120 cgcacaagtc gcaccgcaag
ccgaagcgct cgtggaacgt gtacgtgggc cgctcgctga 180 aggcgatcaa cgcccagatg tcgatgtcgc
accgcacgat gaagatcgtg aactcgtacg 240 tgaacgacgt gatggagcgc atctgcactg aggccgcgtc
gattgttcgc gcgaacaaga 300 agcgcacgtt gggtgcgcgc gaggtgcaga cggcgggtgcg cattgtgctg
ccggcggagc 360 tcgcgaagca tgccattggct gagggcacga aggccgtgtc gaggcgtcc cgctaaagcg
420 gcttgccgga tgccgtgtga gtaggaggtt ggcttgccgc aaacgctgac ctgcgcgatt 480
gcggcgtggc gctccccttc tctccttgt cggcgggtgt gtgtcatgca tttgcgtgac 540 tctcctctct
tatagatgca agcttttttt ttctcttgac gttttatttt ctctccccc 600 tcccttaacg tgaagtgtat
atganagcgt actggacatg ananaaaaaa aaaaanaaact 660 cgag 664 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 73 <211> LENGTH: 1432 <212> TYPE: DNA <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222>
LOCATION: (1)...(1432) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 73
gatgaagaag aggaggacac caccatcaac aactccgacg tgggtggtgcg ctacaagaag 60 gccgcaacgt
ggtgcaatga aacgttgccg gtgcttatcg atgccacaaa acctggcgcc 120 aagggtgtgcg acctgtgccg
cctcgggtgat gacaccatca ccgcnaggt caagacaatg 180 ttcaaaggca cggaaaaagg catcgctttc
ccgacctgca tctcgggtcaa caactgcgta 240 tgccacaaca gccctggcgt gtcggacgag acgacgcagc
aagagatcgc gatgggtgac 300 gtcgtgcact acgacctggg catccacgtg gacggctact gcgccgtcgt
cgcgcacacc 360 attcaggtga cagaggacaa tgagcttggc aaggacgaga aggcggcgcg cgtcattaca
420 gcggcgta acatcctgaa cacggcgctg cgccagatgc gtcccggtag gaccatctac 480
caggtgacag acgtagtga gaaggctgcg gagcactaca aggtgactcc ggtagacggc 540 gtcctctcgc
atatgatgaa gcgctacatc atagacngat accgctgtat cccgcagcgc 600 agggtcgcgcg agcacatggt
gcacgactac gatctcgaga aagcgcaggt gtggacgcta 660 gacattgtca tgacctccg caagggaag
ctgaaggagc gcgatgcgcg gccgtgcgtg 720 ttcaaggtgg ctctggactc caactactct gtgaaaaatg
aaagcgcgaa ggaggttcag 780 aaggaaatcg actccnagta tgccaccttc ccctttgcca tccgcaacct
ggaggccaag 840 aaggccccgc tcggtctcaa cgagatggcg aagcacggtg ctgtcatccc gtacctatt
900 ctcttcgaaa aggaaggcga ggtcgtcgcc catttcaaga ttacggtgct catcagcaac 960
aagaagattg agccgattac cggcctgaag ccgcagaagg ccccgcgct cgagccatac 1020 acggacgaga
tgctgcttgac gacgaacaag ctcttcgctg tcgctagaga agaaggcgcc 1080 gaagtagacg gccgtggcat
ccgtgacgct gtactgcgag ctctcgtagg cgtacgcctc 1140 ttgtgaggcg tacacgtgtg ctgtttgcgg
acgaggaggc acccattctg ttccccttct 1200 tcgctaactt tcgcgtttcc tctgacgctg gcttctytgc
cggagtgtgg tgaggcgcgt 1260 gggggagaaa cggcccacty gcatgcctgt gcatacgca gcacggtagg
gagcgcggtg 1320 tgtgtgtgtg tggggggcg tgttacgagt acaaaagagg ctcgatcttt gcgatcttt
1380 ttttctgtaa acaggaacat aagtaaccaa aaaaaaaaaa aaaaaactcg ag 1432 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 74 <211> LENGTH: 873 <212> TYPE: DNA <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222>
LOCATION: (1)...(873) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 74
ctttattgtc atcactgtaa agcactgttt tttctttcac ttttcttga gtgttttctt 60 ctattcacca
tgagcattat caaggaggac gacgcggtgg gctgctacat gacggtgacc 120 ctgctggacg acaccaaggt
ggagggtacc atcttaccct acaattccaa ggagggcata 180 atagtactcc tgtccctccg cgacgatcag
acgaacatga agtaataccg cactccgtac 240 atcaaagact ctacgctttc acacgtgtag gagggagcgc
acctgcccc ggcactggac 300 tcttcaacgc agcttccgtc catgcacgcc ggccgcgaca agtccatctt
caagcacgcc 360 agcacgcagc tcaagaacgc cgaaggcgaac cgcgaaaagc acttcaactc tgtcacgacc
420 gacacaccga ttgccacact tgatgcgtac ctcaagctcc tgcggctata ccccttaatt 480
gagtgaaca gcgacgaggg tgtcatccag gtctcgga caagcattgt cgtaggagac 540 cccgactggc
ggacgcccc ggcaatgctg gtggacggcg cccctgagaa ggacagaccg 600 cttgtagatc gcctgcaggt
tgcgctcggm aacggcaaga agtgattcag tgtgtagcgg 660 acagaacatc gtgtgcttgt gtgtctgttt

gangtttgtt tgttttctct ttgtggtact 720 gcgtacgacg gcgccttctc ccggtggtgg gtgagttccat
aagcagttga gttctygggt 780 gtagnaavgc ctyacygccg accatatggg agagggcgaa caaatntttg
atagaagttg 840 aaaatcccaa agtyaaaaga aaaaaaaaaa aaa 873 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 75 <211> LENGTH: 1238 <212> TYPE: DNA <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: misc_feature <222>
LOCATION: (1)...(1238) <223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 75
tttctgtact ttattgaaca tcagtagaac acgttcttcc cgcaaagatg gccaaagaagc 60 acctcaagcg
cttgtatgcg cccaaggact ggatgctgag caagctgacc ggcgtgttcg 120 cgccgcgtcc ggcgtccgggt
ccgcacaagc tgcgcgagtg cctgccgctn ctggtgatca 180 tccgcaaccg gctgaagtac ggcgtgaacg
cgcgcgaggg tgagatgac ctgcgccagg 240 gtctggtgca cgtggacaac caccgcgcc ggcagcgcaa
gtatcccgcc ggtttcatgg 300 acgtggtcga gatccgaag acgggcgacc gcttccgcct gatgtacgac
gtcaagggcc 360 gcttcgcgtt ggtgaacctg tccgaggcgg aggcgcagat caagctgatg aaggttgtga
420 acctgtacac ggcaccggc cgcgtgccgg tcgctgtgac gcacgacggc caccgcatcc 480
gctaccggga ccgcacacc tccattggtg acaccatcgt gtacaacgtc aaggagaaga 540 agtgcgtgga
cctgatcaag aaccgccagg gcaaggccgt gatcgtgacc ggtggcgcca 600 accgcggccg catcggcgag
atcgtgaagg tggagtgcca ccccggtgcg ttcaacattg 660 cgcacctgaa ggacgcgtcc ggcgccgagt
tcgccaccgg cgccgcgaac atcttcgtga 720 tcggcaagga cctgaacaac ctgcaggtaa cggtgccgaa
gcagcagggc ctgcgcatga 780 acgtgatcca ggagcgcgag gagcgctga tcgcggcgga ggcccgcaag
aacgcgccgg 840 ctggtggtgc ccgcagggcc cgcaagtga gaggcgatta cacgcatgcg tgtttgtggc
900 tctgaagcga cttggcgggt cggctgtgag ggtttgagag gaggtgtgtg atgcgtgtga 960
agtcccttc cgttctcagc tctctctgtg ctgtagctgt gcctttcccc agatcgcttt 1020 accgcatttg
catcatctg tgtagtcga tgtgcgtgtt tctgtctctc ggtgggtctc 1080 cctctccctc ccttctgccc
tctctctttg agtgggtgtg catgcgtcgc gcgcagcggg 1140 ctccgcttna gtgattctct cgtgttttan
ggctgtttty tttctyagtt nagcgtttty 1200 gttcatgatt tctcagacc caaaaaaaaa aaaaaaaa
1238 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 76 <211> LENGTH: 712 <212>
TYPE: DNA <213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY:
misc_feature <222> LOCATION: (1)...(712) <223> OTHER INFORMATION: n = A,T,C or G
<400> SEQUENCE: 76 ctgacggagt tccagacgaa ccttgtgccg taccgcgca tccacttcgt
gctgacaagc 60 tacgctccgg tgggtgtctg cgagaaggcg taccacgagc agctntccgt cgcggacatc 120
acgaactcgg tntttgagcc tgctggcatg ctnacaaagt gcgacctcg ccacggcaag 180 tacatgtcgt
gctgcctcat gtaccgcgt gatgtcgtg cgaaggatgt caacgcccg 240 attgcgacga tcaagacgaa
gcgcacaatt cagttcgtgg actggtgcc gaccggcttc 300 aagtgcggca tcaactacca gccgccgacc
gttgtgccg gcggtgacct cgcgaagggtg 360 cagcgcggcg tgtgcatgat tgccaactcg accgcgatcg
ctgaggtgtt tgcccgcac 420 gaccacaagt tcgacctgat gtacagcaag cgcgcgtttg tgactggta
cgtgggtgag 480 ggcattggagg agggcgagtt ctccgaggcg cgcgaggatc tcgctgcgct ggagaaggac
540 tacgaggagg ttggcgccga gtccgccgac gacatggcg aggaggacgt cgaggagtac 600
taaggtagac tcgtgcccg cgctgatgat gtaggtcac gcgtgcgtgt gctgcagcg 660 agccgccgcc
accgcgactg tgtgtgtgtg cgcgcgtgac gaccggctcg ag 712 <200> SEQUENCE CHARACTERISTICS:
<210> SEQ ID NO 77 <211> LENGTH: 1086 <212> TYPE: DNA <213> ORGANISM: Leishmania
major <220> FEATURE: <221> NAME/KEY: misc_feature <222> LOCATION: (1)...(1086)
<223> OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 77 caagaagtgg atcaagcagg
agacgaacgc cgatggcgag cgcgtgcgcc gcgcttctg 60 caagtctgc ctgacccca tctaccagat
cttcgacgct gtgatgaacg agaagaagga 120 caaggtggac aagatgctca agtcgctgca cgtgacgctn
acggctgagg agcgcgagca 180 ggtgccgaan aagcttctga agacggtgat gatgaanttc ctgccggctg
ctgagacgct 240 gctacagatg atcgtggcg acctgccgtc gcccagaag gcgcaggcgt accgtgcgga
300 gatgctgtac tctggcgagg cgtgcgccga ggacaagtac ttcattgggtg tcaagaactg 360
cgaccccgct gcgccgtca tgctgtacat cagcaagatg gtgccgacgg ccgaccgcgg 420 ccgcttcttc
gcctttggcc gcattcttc cgtaagggtg gcgagcgcc agaagggtgc 480 catcatgggt aacaactacg
tctacggcaa gaagcaggac ctgtacgagg acaagcctgt 540 gcagcgtcc gtgctgatga tgggccgcta
ccaggaggcc gtggaggaca tgccgtgcgg 600 taacgtggtg ggccttgtgg gcgtggacaa gtacatcgtg
aagtccgcga cgatcacgga 660 cgatggcgag agccgcacc cgctgcgcga catgaagtac tctgtgtcgc
ccgtcgtgcg 720 tgtggccgtg gaggcgaaga acccgccga cctgccgaag cttgtggagg gcctgaagcg
780 ccttgccaag tccgacccgc tgggtggtgt cagcattgag gagtctggcg agcacattgt 840
tgccggcgct ggcgagcttc accttgagat ttgcctgaag gatctccagg aggaattcat 900 gaacggcgcg
ccgctnaaga tctccgagcc ggtggtgtcg ttccgcgaga cggtagcgga 960 tgtgtcgtcg cagcagtgcc
tgtcgaagtc tgcgaacaag cacaaccgtc tcttctgccg 1020 cgtgccccg ctnacagagg anctggcgct
ggcgatngan gaaggcaccg ctggtcccga 1080 ncgga 1086 <200> SEQUENCE CHARACTERISTICS:
<210> SEQ ID NO 78 <211> LENGTH: 447 <212> TYPE: DNA <213> ORGANISM: Leishmania
major <220> FEATURE: <221> NAME/KEY: misc_feature <222> LOCATION: (1)...(447) <223>
OTHER INFORMATION: n = A,T,C or G <400> SEQUENCE: 78 cgcacaaacg tctacttcga
tnagtcgacg ggaggccgct acgtgccggc gcgcgtgctg 60 atggacctcg agccggcac tatggactcc
gttcgcggcg gcccgtagcg ccagctgttc 120 cgcccggaac acttcatctt tggtcagtcc ggcgctggca
acaactgggc caagggccac 180 tacactgagg gcgcggagct gatcgactcc gtgcttgatg tgtgccgcaa
ggaggcgag 240 agctgcgact gcctgcagg cttccagctg tctactccc tcggcgcgcg cacgggctcc

300 ggcattgggca cgctgctcat ttccaanctg cgcgangagt acccggaccg gatcatgatg 360
accttctccg tcatccgctc cccccgctg tcggataccg ttgtggancc gtacaacacg 420 accctctctg
tgcaccagct cgtggaa 447 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 79 <211>
LENGTH: 375 <212> TYPE: DNA <213> ORGANISM: Leishmania major <220> FEATURE: <221>
NAME/KEY: misc_feature <222> LOCATION: (1)...(375) <223> OTHER INFORMATION: n =
A,T,C or G <400> SEQUENCE: 79 gtaacccgct ggtgtacgca tatgtagaca cagacgggca
gcacgagacg acgttctctg 60 cgatccctgt ggtgcttggc atgaatggaa tcgagaagcg cctgccgatt
ggtccgctgc 120 actcgacgga ggaacgctg ctgaaggcgg cactgccggt gatcaagaag aatatcgtga
180 agggcagcga gttcgcgcg ctcacacctgt agcacctcag cttttttttt ttgcgttaaa 240
cgggcgtggg aagcacctcg atacttctgct tcgcgctgac ggacccgcac gacatcgttc 300 gtcatcccc
tccccctctt cggccctata cgcatgaagg agtggaaatta tgcaacagca 360 tgtnnatatc aagtg 375
<200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 80 <211> LENGTH: 107 <212> TYPE:
PRT <213> ORGANISM: Leishmania major <400> SEQUENCE: 80 Met Ala Ser Ser Arg Lys Ala
Ser Asn Pro His Lys Ser His Arg Lys 1 5 10 15 Pro Lys Arg Ser Trp Asn Val Tyr Val
Gly Arg Ser Leu Lys Ala Ile 20 25 30 Asn Ala Gln Met Ser Met Ser His Arg Thr Met
Lys Ile Val Asn Ser 35 40 45 Tyr Val Asn Asp Val Met Glu Arg Ile Cys Thr Glu Ala
Ala Ser Ile 50 55 60 Val Arg Ala Asn Lys Lys Arg Thr Leu Gly Ala Arg Glu Val Gln
Thr 65 70 75 80 Ala Val Arg Ile Val Leu Pro Ala Glu Leu Ala Lys His Ala Met Ala 85
90 95 Glu Gly Thr Lys Ala Val Ser Ser Ala Ser Arg 100 105 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 81 <211> LENGTH: 381 <212> TYPE: PRT <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION:
(1)...(381) <223> OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 81 Asp
Glu Glu Glu Glu Asp Thr Thr Ile Asn Asn Ser Asp Val Val Val 1 5 10 15 Arg Tyr Lys
Lys Ala Ala Thr Trp Cys Asn Glu Thr Leu Arg Val Leu 20 25 30 Ile Asp Ala Thr Lys
Pro Gly Ala Lys Val Cys Asp Leu Cys Arg Leu 35 40 45 Gly Asp Asp Thr Ile Thr Ala
Xaa Val Lys Thr Met Phe Lys Gly Thr 50 55 60 Glu Lys Gly Ile Ala Phe Pro Thr Cys
Ile Ser Val Asn Asn Cys Val 65 70 75 80

Detailed Description Paragraph Table (12):

Cys His Asn Ser Pro Gly Val Ser Asp Glu Thr Thr Gln Gln Glu Ile 85 90 95 Ala Met
Gly Asp Val Val His Tyr Asp Leu Gly Ile His Val Asp Gly 100 105 110 Tyr Cys Ala Val
Val Ala His Thr Ile Gln Val Thr Glu Asp Asn Glu 115 120 125 Leu Gly Lys Asp Glu Lys
Ala Ala Arg Val Ile Thr Ala Ala Tyr Asn 130 135 140 Ile Leu Asn Thr Ala Leu Arg Gln
Met Arg Pro Gly Thr Thr Ile Tyr 145 150 155 160 Gln Val Thr Asp Val Val Glu Lys Ala
Ala Glu His Tyr Lys Val Thr 165 170 175 Pro Val Asp Gly Val Leu Ser His Met Met Lys
Arg Tyr Ile Ile Asp 180 185 190 Xaa Tyr Arg Cys Ile Pro Gln Arg Arg Val Ala Glu His
Met Val His 195 200 205 Asp Tyr Asp Leu Glu Lys Ala Gln Val Trp Thr Leu Asp Ile Val
Met 210 215 220 Thr Ser Gly Lys Gly Lys Leu Lys Glu Arg Asp Ala Arg Pro Cys Val 225
230 235 240 Phe Lys Val Ala Leu Asp Ser Asn Tyr Ser Val Lys Met Glu Ser Ala 245 250
255 Lys Glu Val Gln Lys Glu Ile Asp Ser Xaa Tyr Ala Thr Phe Pro Phe 260 265 270 Ala
Ile Arg Asn Leu Glu Ala Lys Lys Ala Arg Leu Gly Leu Asn Glu 275 280 285 Met Ala Lys
His Gly Ala Val Ile Pro Tyr Pro Ile Leu Phe Glu Lys 290 295 300 Glu Gly Glu Val Val
Ala His Phe Lys Ile Thr Val Leu Ile Ser Asn 305 310 315 320 Lys Lys Ile Glu Pro Ile
Thr Gly Leu Lys Pro Gln Lys Ala Pro Ala 325 330 335 Leu Glu Pro Tyr Thr Asp Glu Met
Leu Leu Ala Thr Asn Lys Leu Phe 340 345 350 Ala Val Ala Arg Glu Glu Gly Gly Glu Val
Asp Gly Arg Gly Ile Arg 355 360 365 Asp Ala Val Leu Arg Ala Phe Val Gly Val Arg Leu
Leu 370 375 380 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 82 <211> LENGTH:
191 <212> TYPE: PRT <213> ORGANISM: Leishmania major <400> SEQUENCE: 82 Met Ser Ile
Ile Lys Glu Asp Asp Ala Val Gly Cys Tyr Met Thr Val 1 5 10 15 Thr Leu Val Asp Asp
Thr Lys Val Glu Gly Thr Ile Phe Thr Tyr Asn 20 25 30 Ser Lys Glu Gly Ile Ile Val
Leu Leu Ser Leu Arg Asp Asp Gln Thr 35 40 45 Asn Met Lys Leu Ile Arg Thr Pro Tyr
Ile Lys Asp Phe Ser Leu Ser 50 55 60 His Ala Glu Glu Gly Ala His Leu Pro Pro Ala
Leu Asp Ser Phe Asn 65 70 75 80 Glu Leu Pro Ser Met His Ala Gly Arg Asp Lys Ser Ile
Phe Lys His 85 90 95 Ala Ser Thr Gln Leu Lys Asn Ala Glu Ala Asn Arg Glu Lys His
Phe 100 105 110 Asn Ser Val Thr Thr Asp Thr Pro Ile Ala Thr Leu Asp Ala Tyr Leu 115
120 125 Lys Leu Leu Arg Leu Tyr Pro Leu Ile Glu Trp Asn Ser Asp Glu Gly 130 135 140
Val Ile Gln Val Ser Asp Thr Val Ile Val Val Gly Asp Pro Asp Trp 145 150 155 160 Arg
Thr Pro Lys Ala Met Leu Val Asp Gly Ala Pro Glu Lys Asp Arg 165 170 175 Pro Leu Val
Asp Arg Leu Gln Val Ala Leu Gly Asn Gly Lys Lys 180 185 190 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 83 <211> LENGTH: 273 <212> TYPE: PRT <213>
ORGANISM: Leishmania major <400> SEQUENCE: 83 Met Ala Lys Lys His Leu Lys Arg Leu
Tyr Ala Pro Lys Asp Trp Met 1 5 10 15 Leu Ser Lys Leu Thr Gly Val Phe Ala Pro Arg
Pro Arg Pro Gly Pro 20 25 30 His Lys Leu Arg Glu Cys Leu Pro Leu Leu Val Ile Ile
Arg Asn Arg 35 40 45 Leu Lys Tyr Ala Leu Asn Ala Arg Glu Gly Glu Met Ile Leu Arg

Gln 50 55 60 Gly Leu Val His Val Asp Asn His Pro Arg Arg Asp Gly Lys Tyr Pro 65 70
75 80 Ala Gly Phe Met Asp Val Val Glu Ile Pro Lys Thr Gly Asp Arg Phe 85 90 95 Arg
Leu Met Tyr Asp Val Lys Gly Arg Phe Ala Leu Val Asn Leu Ser 100 105 110 Glu Ala Glu
Ala Gln Ile Lys Leu Met Lys Val Val Asn Leu Tyr Thr 115 120 125 Ala Thr Gly Arg Val
Pro Val Ala Val Thr His Asp Gly His Arg Ile 130 135 140 Arg Tyr Pro Asp Pro His Thr
Ser Ile Gly Asp Thr Ile Val Tyr Asn 145 150 155 160 Val Lys Glu Lys Lys Cys Val Asp
Leu Ile Lys Asn Arg Gln Gly Lys 165 170 175 Ala Val Ile Val Thr Gly Gly Ala Asn Arg
Gly Arg Ile Gly Glu Ile 180 185 190 Val Lys Val Glu Cys His Pro Gly Ala Phe Asn Ile
Ala His Leu Lys 195 200 205 Asp Ala Ser Gly Ala Glu Phe Ala Thr Arg Ala Ala Asn Ile
Phe Val 210 215 220 Ile Gly Lys Asp Leu Asn Asn Leu Gln Val Thr Val Pro Lys Gln Gln
225 230 235 240 Gly Leu Arg Met Asn Val Ile Gln Glu Arg Glu Glu Arg Leu Ile Ala 245
250 255 Ala Glu Ala Arg Lys Asn Ala Pro Ala Arg Gly Ala Arg Arg Ala Arg 260 265 270
Lys <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 84 <211> LENGTH: 200 <212>
TYPE: PRT <213> ORGANISM: Leishmania major <400> SEQUENCE: 84 Leu Thr Glu Phe Gln
Thr Asn Leu Val Pro Tyr Pro Arg Ile His Phe 1 5 10 15 Val Leu Thr Ser Tyr Ala Pro
Val Val Ser Ala Glu Lys Ala Tyr His 20 25 30 Glu Gln Leu Ser Val Ala Asp Ile Thr
Asn Ser Val Phe Glu Pro Ala 35 40 45 Gly Met Leu Thr Lys Cys Asp Pro Arg His Gly
Lys Tyr Met Ser Cys 50 55 60 Cys Leu Met Tyr Arg Gly Asp Val Val Pro Lys Asp Val
Asn Ala Ala 65 70 75 80 Ile Ala Thr Ile Lys Thr Lys Arg Thr Ile Gln Phe Val Asp Trp
Cys 85 90 95 Pro Thr Gly Phe Lys Cys Gly Ile Asn Tyr Gln Pro Pro Thr Val Val 100
105 110 Pro Gly Gly Asp Leu Ala Lys Val Gln Arg Ala Val Cys Met Ile Ala 115 120 125
Asn Ser Thr Ala Ile Ala Glu Val Phe Ala Arg Ile Asp His Lys Phe 130 135 140 Asp Leu
Met Tyr Ser Lys Arg Ala Phe Val His Trp Tyr Val Gly Glu 145 150 155 160 Gly Met Glu
Glu Gly Glu Phe Ser Glu Ala Arg Glu Asp Leu Ala Ala 165 170 175 Leu Glu Lys Asp Tyr
Glu Glu Val Gly Ala Glu Ser Ala Asp Asp Met 180 185 190 Gly Glu Glu Asp Val Glu Glu
Tyr 195 200 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 85 <211> LENGTH: 361
<212> TYPE: PRT <213> ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY:
VARIANT <222> LOCATION: (1)...(361) <223> OTHER INFORMATION: Xaa = Any Amino Acid
<400> SEQUENCE: 85 Lys Lys Trp Ile Lys Gln Glu Thr Asn Ala Asp Gly Glu Arg Val Arg
1 5 10 15 Arg Ala Phe Cys Gln Phe Cys Leu Asp Pro Ile Tyr Gln Ile Phe Asp 20 25 30
Ala Val Met Asn Glu Lys Lys Asp Lys Val Asp Lys Met Leu Lys Ser 35 40 45 Leu His
Val Thr Leu Thr Ala Glu Glu Arg Glu Gln Val Pro Xaa Lys 50 55 60 Leu Leu Lys Thr
Val Met Met Xaa Phe Leu Pro Ala Ala Glu Thr Leu 65 70 75 80 Leu Gln Met Ile Val Ala
His Leu Pro Ser Pro Lys Lys Ala Gln Ala 85 90 95 Tyr Arg Ala Glu Met Leu Tyr Ser
Gly Glu Ala Ser Pro Glu Asp Lys 100 105 110 Tyr Phe Met Gly Ile Lys Asn Cys Asp Pro
Ala Ala Pro Leu Met Leu 115 120 125 Tyr Ile Ser Lys Met Val Pro Thr Ala Asp Arg Gly
Arg Phe Phe Ala 130 135 140 Phe Gly Arg Ile Phe Ser Gly Lys Val Arg Ser Gly Gln Lys
Val Arg 145 150 155 160 Ile Met Gly Asn Asn Tyr Val Tyr Gly Lys Lys Gln Asp Leu Tyr
Glu 165 170 175 Asp Lys Pro Val Gln Arg Ser Val Leu Met Met Gly Arg Tyr Gln Glu 180
185 190 Ala Val Glu Asp Met Pro Cys Gly Asn Val Val Gly Leu Val Gly Val 195 200 205
Asp Lys Tyr Ile Val Lys Ser Ala Thr Ile Thr Asp Asp Gly Glu Ser 210 215 220 Pro His
Pro Leu Arg Asp Met Lys Tyr Ser Val Ser Pro Val Val Arg 225 230 235 240 Val Ala Val
Glu Ala Lys Asn Pro Ser Asp Leu Pro Lys Leu Val Glu 245 250 255 Gly Leu Lys Arg Leu
Ala Lys Ser Asp Pro Leu Val Val Cys Ser Ile 260 265 270 Glu Glu Ser Gly Glu His Ile
Val Ala Gly Ala Gly Glu Leu His Leu 275 280 285 Glu Ile Cys Leu Lys Asp Leu Gln Glu
Asp Phe Met Asn Gly Ala Pro 290 295 300 Leu Lys Ile Ser Glu Pro Val Val Ser Phe Arg
Glu Thr Val Thr Asp 305 310 315 320 Val Ser Ser Gln Gln Cys Leu Ser Lys Ser Ala Asn
Lys His Asn Arg 325 330 335 Leu Phe Cys Arg Gly Ala Pro Leu Thr Glu Xaa Leu Ala Leu
Ala Xaa 340 345 350 Xaa Glu Gly Thr Ala Gly Pro Xaa Ala 355 360 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 86 <211> LENGTH: 149 <212> TYPE: PRT <213>
ORGANISM: Leishmania major <220> FEATURE: <221> NAME/KEY: VARIANT <222> LOCATION:
(1)...(149) <223> OTHER INFORMATION: Xaa = Any Amino Acid <400> SEQUENCE: 86 Arg
Ile Asn Val Tyr Phe Asp Xaa Ser Thr Gly Gly Arg Tyr Val Pro 1 5 10 15 Arg Ala Val
Leu Met Asp Leu Glu Pro Gly Thr Met Asp Ser Val Arg 20 25 30 Ala Gly Pro Tyr Gly
Gln Leu Phe Arg Pro Asp Asn Phe Ile Phe Gly 35 40 45 Gln Ser Gly Ala Gly Asn Asn
Trp Ala Lys Gly His Tyr Thr Glu Gly 50 55 60 Ala Glu Leu Ile Asp Ser Val Leu Asp
Val Cys Arg Lys Glu Ala Glu 65 70 75 80 Ser Cys Asp Cys Leu Gln Gly Phe Gln Leu Ser
His Ser Leu Gly Gly 85 90 95 Gly Thr Gly Ser Gly Met Gly Thr Leu Leu Ile Ser Xaa
Leu Arg Xaa 100 105 110 Glu Tyr Pro Asp Arg Ile Met Met Thr Phe Ser Val Ile Pro Ser
Pro 115 120 125 Arg Val Ser Asp Thr Val Val Xaa Pro Tyr Asn Thr Thr Leu Ser Val 130
135 140 His Gln Leu Val Glu 145 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 87
<211> LENGTH: 69 <212> TYPE: PRT <213> ORGANISM: Leishmania major <400> SEQUENCE:
87 Asn Pro Leu Val Tyr Ala Tyr Val Asp Thr Asp Gly Gln His Glu Thr 1 5 10 15 Thr
Phe Leu Ala Ile Pro Val Val Leu Gly Met Asn Gly Ile Glu Lys 20 25 30 Arg Leu Pro

Ile Gly Pro Leu His Ser Thr Glu Glu Thr Leu Leu Lys 35 40 45 Ala Ala Leu Pro Val
 Ile Lys Lys Asn Ile Val Lys Gly Ser Glu Phe 50 55 60 Ala Arg Ser His Leu 65 <200>
 SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 88 <211> LENGTH: 54 <212> TYPE: DNA <213>
 ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR primer
 <400> SEQUENCE: 88

Detailed Description Paragraph Table (13):

agtattcata tgcaccacca ccaccaccac atgtcctgcg gtaacgcca gatc 54 <200> SEQUENCE
 CHARACTERISTICS: <210> SEQ ID NO 89 <211> LENGTH: 33 <212> TYPE: DNA <213>
 ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR primer
 <400> SEQUENCE: 89 ctcacaggat ccctgcttgc tgaagtatcc ttc 33 <200> SEQUENCE
 CHARACTERISTICS: <210> SEQ ID NO 90 <211> LENGTH: 36 <212> TYPE: DNA <213>
 ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR primer
 <400> SEQUENCE: 90 catttcggat ccatggacgc aactgagctg aagaac 36 <200> SEQUENCE
 CHARACTERISTICS: <210> SEQ ID NO 91 <211> LENGTH: 33 <212> TYPE: DNA <213>
 ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR primer
 <400> SEQUENCE: 91 cgtagagaat tcctgaccaa aacgaatgat gcc 33 <200> SEQUENCE
 CHARACTERISTICS: <210> SEQ ID NO 92 <211> LENGTH: 33 <212> TYPE: DNA <213>
 ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR primer
 <400> SEQUENCE: 92 caccacgaat tcatggcgca gaatgataag atc 33 <200> SEQUENCE
 CHARACTERISTICS: <210> SEQ ID NO 93 <211> LENGTH: 34 <212> TYPE: DNA <213>
 ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: PCR primer
 <400> SEQUENCE: 93 actgacctcg aggaattctt agtcgcgcac gaac 34 <200> SEQUENCE
 CHARACTERISTICS: <210> SEQ ID NO 94 <211> LENGTH: 3012 <212> TYPE: DNA <213>
 ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: DNA sequence
 encoding fusion (poly-protein) constructs comprising multiple Leishmania antigens
 <400> SEQUENCE: 94 catatgcacc accaccacca ccacatgtcc tgcggtaacg ccaagatcaa
 ctctcccgcg 60 ccgtccttcg aggaggtggc gctcatgccc aacggcagct tcaagaagat cagcctctcc 120
 tcctacaagg gcaagtgggt cgtgctcttc ttctacccgc tcgacttcac cttcgtgtgc 180 ccgacagagg
 tcatcgctt ctcgacagc gtgagtcgt tcaacgagct caactgcgag 240 gtcctcgct gctcgataga
 cagcgagtac gcgcacctgc agtgagcgt gcaggaccgc 300 aagaagggcg gcctcgggac catggcgatc
 ccaatgctag ccgacaagac caagagcatc 360 gtcgttcct acggcgtgct ggaggagagc cagggcgtgg
 cctaccgcgg tctcttcac 420 atcgaccccc atggcatgct gcgtcagatc accgtcaatg acatgccggt
 gggccgcagc 480 gtggaggagg ttctacgcct gctggaggct tttcagttcg tggagaagca cggcgagggtg
 540 tgccccgcga actggaagaa gggcgcccc acgatgaagc cggaaccgaa tgcgtctgtc 600
 gagggatact tcagcaagca gggatccatg gacgcaactg agctgaagaa caaggggaac 660 gaagagttct
 ccgcccggcg ctatgtggag gcggtgaact acttctcaaa ggcatccag 720 ttggatgagc agaacagtgt
 cctctacagc aaccgctccg cctgttttgc agccatgcag 780 aaatacaagg acgcgctgga cgacccgac
 aagtgcatt cgatcaagcc gaattgggcc 840 aagggtacg tgcgccgagg agcagctctc catggcatgc
 gccgtacga cgatgccatt 900 gccgcgtatg aaaaggggct caaggtggac cttccaaca gcggtgcgc
 gcaggcgctg 960 aaggacgtgc aggtagcaa gggccgcgaa gcacgtgacc ccacgtctcg cgtcttcacc
 1020 ccgagggcgt tccgcaagat ccaagagaat ccaagctgt ctctacttat gtcgcagccg 1080
 gactacgtga agatggtaga caccgtcatc gcgcaccctt cgcagggccg gctgtacatg 1140 gaagaccagc
 gctttgccct gacgtcatg tacctgagcg gaatgaagat tcccaacgat 1200 ggtgatggcg aggaggagga
 acgtccgtct gcgaaggcgg cagagacagc gaagccaaaa 1260 gaggagaagc ctctcaccga caacgagaag
 gaggccctgg cgctcaagga ggagggaac 1320 aagctgtacc tctcgaagaa gtttgaggag gcgctgacca
 agtaccaaga ggccaggtg 1380 aaagacccca acaacacttt atacattctg aacgtgtcgg ccgtgtactt
 cgagcagggt 1440 gactacgaca agtgcacgc cgagtgcgag caggtatcg agcacggctc cgagaaccac
 1500 tgcgactaca caatcattgc gaagctcatg acccggaacg ctttgtgcct ccagaggcag 1560
 aggaagtacg aggtgctat cgacctttac aagcgcgccc ttgtcagtg gcgtaaccct 1620 gacaccctca
 agaagctgac ggagtgcgag aaggagcacc aaaaggcggt ggagggaagc 1680 tacatcgatc ctgagatcgc
 gaagcagaag aaagacgaag gtaaccagta cttcaaggag 1740 gataagttcc ccgaggccgt ggcagcgtac
 acggaggcca tcaagcgcaa ccctgccgag 1800 cacacctcct acagcaatcg cgcggccgcg tacatcaagc
 ttggagcctt caacgacgcc 1860 ctcaaggacg cgagaagtg cattgagctg aagcccgact ttgttaaggg
 ctacgcgcgc 1920 aagggtcatg cttacttttg gaccaagcag tacaaccgcg cgctgcaggc gtacaatgag
 1980 ggcctcaagg tggaccgag caatgcggac tgcaaggatg ggcggtatcg cacaatcatg 2040
 aagattcagg agatggcatc tggccaatcc gcggtggcg acgaggcggc gcgccgggccc 2100 atggacgatc
 ctgaaatcgc ggcaatcatg caagatagct acatgcaact agtgttgaag 2160 gagatgcaga acgatccac
 gcgcattcag gactacatga aggactccg gatctcatcg 2220 aagatcaaca agctgatttc agctggcatc
 attcgttttg gtcaggaatt catggcgag 2280 aatgataaga tcgccccca ggaccaggac tccttcctcg
 atgaccagcc cggcgttcgc 2340 ccgatccgt ctttcgacga catgccgctg caccagaacc tgctgcgtgg
 catctactcg 2400 tacgggttcg agaagccgtc cagcatccag cagcgcgcga tagccccctt cagcgcggc
 2460 ggcgacatca tcgcgcaggc ccagtccgt accggcaaga cgggtgcctt ctccatcggt 2520
 ctgctgcagc gcttgactt ccgccacaac ctgatccagg gctcgtgct ctccccact 2580 cgcgagctgg

ccctgcagac ggcggaggtg atcagccgca tcggtgagtt cctgtcgaac 2640 agctccaagt tctgcgagac
ctttgtcggc ggcacgcgcg tgcaggatga cctgcgcaag 2700 ctgcaggccg gcgtcatcgt tgccgtgggc
acgccggggc gcgtgtccga cgtgatcaag 2760 cgtggcgcgc tgcgcacaga gtcgtgcgcg gtgctgggtgc
tcgacgagggc tgatgagatg 2820 ctgtctcagg gcttcgcgga ccagatttac gagatcttcc gcttctctgcc
gaaggacatc 2880 caggtcgcgc tcttctccgc cactgatgccg gaggaggtac tggagctgac gaagaagtgc
2940 atgcgcgact aagaattcct cgagcagatc cggctgctaa caaagcccga aaggaagctg 3000
aatggctgct gc 3012 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 95 <211> LENGTH:
982 <212> TYPE: PRT <213> ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER
INFORMATION: Fusion (poly-protein) constructs comprising multiple Leishmania
antigens <400> SEQUENCE: 95 Met His His His His His His Met Ser Cys Gly Asn Ala Lys
Ile Asn 5 10 15 Ser Pro Ala Pro Ser Phe Glu Glu Val Ala Leu Met Pro Asn Gly Ser 20
25 30 Phe Lys Lys Ile Ser Leu Ser Ser Tyr Lys Gly Lys Trp Val Val Leu 35 40 45 Phe
Phe Tyr Pro Leu Asp Phe Thr Phe Val Cys Pro Thr Glu Val Ile 50 55 60 Ala Phe Ser
Asp Ser Val Ser Arg Phe Asn Glu Leu Asn Cys Glu Val 65 70 75 80 Leu Ala Cys Ser Ile
Asp Ser Glu Tyr Ala His Leu Gln Trp Thr Leu 85 90 95 Gln Asp Arg Lys Lys Gly Gly
Leu Gly Thr Met Ala Ile Pro Met Leu 100 105 110 Ala Asp Lys Thr Lys Ser Ile Ala Arg
Ser Tyr Gly Val Leu Glu Glu 115 120 125 Ser Gln Gly Val Ala Tyr Arg Gly Leu Phe Ile
Ile Asp Pro His Gly 130 135 140 Met Leu Arg Gln Ile Thr Val Asn Asp Met Pro Val Gly
Arg Ser Val 145 150 155 160 Glu Glu Val Leu Arg Leu Leu Glu Ala Phe Gln Phe Val Glu
Lys His 165 170 175 Gly Glu Val Cys Pro Ala Asn Trp Lys Lys Gly Ala Pro Thr Met Lys
180 185 190 Pro Glu Pro Asn Ala Ser Val Glu Gly Tyr Phe Ser Lys Gln Gly Ser 195 200
205 Met Asp Ala Thr Glu Leu Lys Asn Lys Gly Asn Glu Glu Phe Ser Ala 210 215 220 Gly
Arg Tyr Val Glu Ala Val Asn Tyr Phe Ser Lys Ala Ile Gln Leu 225 230 235 240 Asp Glu
Gln Asn Ser Val Leu Tyr Ser Asn Arg Ser Ala Cys Phe Ala 245 250 255 Ala Met Gln Lys
Tyr Lys Asp Ala Leu Asp Asp Ala Asp Lys Cys Ile 260 265 270 Ser Ile Lys Pro Asn Trp
Ala Lys Gly Tyr Val Arg Arg Gly Ala Ala 275 280 285 Leu His Gly Met Arg Arg Tyr Asp
Asp Ala Ile Ala Ala Tyr Glu Lys 290 295 300 Gly Leu Lys Val Asp Pro Ser Asn Ser Gly
Cys Ala Gln Gly Val Lys 305 310 315 320 Asp Val Gln Val Ala Lys Ala Arg Glu Ala Arg
Asp Pro Ile Ala Arg 325 330 335 Val Phe Thr Pro Glu Ala Phe Arg Lys Ile Gln Glu Asn
Pro Lys Leu 340 345 350 Ser Leu Leu Met Leu Gln Pro Asp Tyr Val Lys Met Val Asp Thr
Val 355 360 365 Ile Arg Asp Pro Ser Gln Gly Arg Leu Tyr Met Glu Asp Gln Arg Phe 370
375 380 Ala Leu Thr Leu Met Tyr Leu Ser Gly Met Lys Ile Pro Asn Asp Gly 385 390 395
400 Asp Gly Glu Glu Glu Glu Arg Pro Ser Ala Lys Ala Ala Glu Thr Ala 405 410 415 Lys
Pro Lys Glu Glu Lys Pro Leu Thr Asp Asn Glu Lys Glu Ala Leu 420 425 430 Ala Leu Lys
Glu Glu Gly Asn Lys Leu Tyr Leu Ser Lys Lys Phe Glu 435 440 445 Glu Ala Leu Thr Lys
Tyr Gln Glu Ala Gln Val Lys Asp Pro Asn Asn 450 455 460 Thr Leu Tyr Ile Leu Asn Val
Ser Ala Val Tyr Phe Glu Gln Gly Asp 465 470 475 480 Tyr Asp Lys Cys Ile Ala Glu Cys
Glu His Gly Ile Glu His Gly Arg 485 490 495 Glu Asn His Cys Asp Tyr Thr Ile Ile Ala
Lys Leu Met Thr Arg Asn 500 505 510 Ala Leu Cys Leu Gln Arg Gln Arg Lys Tyr Glu Ala
Ala Ile Asp Leu 515 520 525 Tyr Lys Arg Ala Leu Val Glu Trp Arg Asn Pro Asp Thr Leu
Lys Lys 530 535 540 Leu Thr Glu Cys Glu Lys Glu His Gln Lys Ala Val Glu Glu Ala Tyr
545 550 555 560 Ile Asp Pro Glu Ile Ala Lys Gln Lys Lys Asp Glu Gly Asn Gln Tyr 565
570 575 Phe Lys Glu Asp Lys Phe Pro Glu Ala Val Ala Ala Tyr Thr Glu Ala 580 585 590
Ile Lys Arg Asn Pro Ala Glu His Thr Ser Tyr Ser Asn Arg Ala Ala 595 600 605 Ala Tyr
Ile Lys Leu Gly Ala Phe Asn Asp Ala Leu Lys Asp Ala Glu 610 615 620 Lys Cys Ile Glu
Leu Lys Pro Asp Phe Val Lys Gly Tyr Ala Arg Lys 625 630 635 640 Gly His Ala Tyr Phe
Trp Thr Lys Gln Tyr Asn Arg Ala Leu Gln Ala 645 650 655 Tyr Asn Glu Gly Leu Lys Val
Asp Pro Ser Asn Ala Asp Cys Lys Asp 660 665 670 Gly Arg Tyr Arg Thr Ile Met Lys Ile
Gln Glu Met Ala Ser Gly Gln 675 680 685 Ser Ala Asp Gly Asp Glu Ala Ala Arg Arg Ala
Met Asp Asp Pro Glu 690 695 700 Ile Ala Ala Ile Met Gln Asp Ser Tyr Met Gln Leu Val
Leu Lys Glu 705 710 715 720 Met Gln Asn Asp Pro Thr Arg Ile Gln Glu Tyr Met Lys Asp
Ser Gly 725 730 735 Ile Ser Ser Lys Ile Asn Lys Leu Ile Ser Ala Gly Ile Ile Arg Phe
740 745 750 Gly Gln Glu Phe Met Ala Gln Asn Asp Lys Ile Ala Pro Gln Asp Gln 755 760
765 Asp Ser Phe Leu Asp Asp Gln Pro Gly Val Arg Pro Ile Pro Ser Phe 770 775 780 Asp
Asp Met Pro Leu His Gln Asn Leu Leu Arg Gly Ile Tyr Ser Tyr 785 790 795 800 Gly Phe
Glu Lys Pro Ser Ser Ile Gln Gln Arg Ala Ile Ala Pro Phe 805 810 815 Thr Arg Gly Gly
Asp Ile Ile Ala Gln Ala Gln Ser Gly Thr Gly Lys 820 825 830 Thr Gly Ala Phe Ser Ile
Gly Leu Leu Gln Arg Leu Asp Phe Arg His 835 840 845 Asn Leu Ile Gln Gly Leu Val Leu
Ser Pro Thr Arg Glu Leu Ala Leu 850 855 860 Gln Thr Ala Glu Val Ile Ser Arg Ile Gly
Glu Phe Leu Ser Asn Ser 865 870 875 880 Ser Lys Phe Cys Glu Thr Phe Val Gly Gly Thr
Arg Val Gln Asp Asp 885 890 895 Leu Arg Lys Leu Gln Ala Gly Val Ile Val Ala Val Gly
Thr Pro Gly 900 905 910 Arg Val Ser Asp Val Ile Lys Arg Gly Ala Leu Arg Thr Glu Ser
Leu 915 920 925 Arg Val Leu Val Leu Asp Glu Ala Asp Glu Met Leu Ser Gln Gly Phe 930
935 940 Ala Asp Gln Ile Tyr Glu Ile Phe Arg Phe Leu Pro Lys Asp Ile Gln 945 950 955

960 Val Ala Leu Phe Ser Ala Thr Met Pro Glu Glu Val Leu Glu Leu Thr 965 970 975 Lys
 Lys Phe Met Arg Asp 980 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 96 <211>
 LENGTH: 1641 <212> TYPE: PRT <213> ORGANISM: Artificial Sequence <220> FEATURE:
 <223> OTHER INFORMATION: Fusion (poly-protein) constructs comprising multiple
Leishmania antigens <400> SEQUENCE: 96 Met His His His His His His Met Ser Cys Gly
 Asn Ala Lys Ile Asn 5 10 15 Ser Pro Ala Pro Ser Phe Glu Glu Val Ala Leu Met Pro Asn
 Gly Ser

Detailed Description Paragraph Table (14):

20 25 30 Phe Lys Lys Ile Ser Leu Ser Ser Tyr Lys Gly Lys Trp Val Val Leu 35 40 45
 Phe Phe Tyr Pro Leu Asp Phe Thr Phe Val Cys Pro Thr Glu Val Ile 50 55 60 Ala Phe
 Ser Asp Ser Val Ser Arg Phe Asn Glu Leu Asn Cys Glu Val 65 70 75 80 Leu Ala Cys Ser
 Ile Asp Ser Glu Tyr Ala His Leu Gln Trp Thr Leu 85 90 95 Gln Asp Arg Lys Lys Gly
 Gly Leu Gly Thr Met Ala Ile Pro Met Leu 100 105 110 Ala Asp Lys Thr Lys Ser Ile Ala
 Arg Ser Tyr Gly Val Leu Glu Glu 115 120 125 Ser Gln Gly Val Ala Tyr Arg Gly Leu Phe
 Ile Ile Asp Pro His Gly 130 135 140 Met Leu Arg Gln Ile Thr Val Asn Asp Met Pro Val
 Gly Arg Ser Val 145 150 155 160 Glu Glu Val Leu Arg Leu Leu Glu Ala Phe Gln Phe Val
 Glu Lys His 165 170 175 Gly Glu Val Cys Pro Ala Asn Trp Lys Lys Gly Ala Pro Thr Met
 Lys 180 185 190 Pro Glu Pro Asn Ala Ser Val Glu Gly Tyr Phe Ser Lys Gln Gly Ser 195
 200 205 Met Asp Ala Thr Glu Leu Lys Asn Lys Gly Asn Glu Glu Phe Ser Ala 210 215 220
 Gly Arg Tyr Val Glu Ala Val Asn Tyr Phe Ser Lys Ala Ile Gln Leu 225 230 235 240 Asp
 Glu Gln Asn Ser Val Leu Tyr Ser Asn Arg Ser Ala Cys Phe Ala 245 250 255 Ala Met Gln
 Lys Tyr Lys Asp Ala Leu Asp Asp Ala Asp Lys Cys Ile 260 265 270 Ser Ile Lys Pro Asn
 Trp Ala Lys Gly Tyr Val Arg Arg Gly Ala Ala 275 280 285 Leu His Gly Met Arg Arg Tyr
 Asp Asp Ala Ile Ala Ala Tyr Glu Lys 290 295 300 Gly Leu Lys Val Asp Pro Ser Asn Ser
 Gly Cys Ala Gln Gly Val Lys 305 310 315 320 Asp Val Gln Val Ala Lys Ala Arg Glu Ala
 Arg Asp Pro Ile Ala Arg 325 330 335 Val Phe Thr Pro Glu Ala Phe Arg Lys Ile Gln Glu
 Asn Pro Lys Leu 340 345 350 Ser Leu Leu Met Leu Gln Pro Asp Tyr Val Lys Met Val Asp
 Thr Val 355 360 365 Ile Arg Asp Pro Ser Gln Gly Arg Leu Tyr Met Glu Asp Gln Arg Phe
 370 375 380 Ala Leu Thr Leu Met Tyr Leu Ser Gly Met Lys Ile Pro Asn Asp Gly 385 390
 395 400 Asp Gly Glu Glu Glu Glu Arg Pro Ser Ala Lys Ala Ala Glu Thr Ala 405 410 415
 Lys Pro Lys Glu Glu Lys Pro Leu Thr Asp Asn Glu Lys Glu Ala Leu 420 425 430 Ala Leu
 Lys Glu Glu Gly Asn Lys Leu Tyr Leu Ser Lys Lys Phe Glu 435 440 445 Glu Ala Leu Thr
 Lys Tyr Gln Glu Ala Gln Val Lys Asp Pro Asn Asn 450 455 460 Thr Leu Tyr Ile Leu Asn
 Val Ser Ala Val Tyr Phe Glu Gln Gly Asp 465 470 475 480 Tyr Asp Lys Cys Ile Ala Glu
 Cys Glu His Gly Ile Glu His Gly Arg 485 490 495 Glu Asn His Cys Asp Tyr Thr Ile Ile
 Ala Lys Leu Met Thr Arg Asn 500 505 510 Ala Leu Cys Leu Gln Arg Gln Arg Lys Tyr Glu
 Ala Ala Ile Asp Leu 515 520 525 Tyr Lys Arg Ala Leu Val Glu Trp Arg Asn Pro Asp Thr
 Leu Lys Lys 530 535 540 Leu Thr Glu Cys Glu Lys Glu His Gln Lys Ala Val Glu Glu Ala
 Tyr 545 550 555 560 Ile Asp Pro Glu Ile Ala Lys Gln Lys Lys Asp Glu Gly Asn Gln Tyr
 565 570 575 Phe Lys Glu Asp Lys Phe Pro Glu Ala Val Ala Tyr Thr Glu Ala 580 585
 590 Ile Lys Arg Asn Pro Ala Glu His Thr Ser Tyr Ser Asn Arg Ala Ala 595 600 605 Ala
 Tyr Ile Lys Leu Gly Ala Phe Asn Asp Ala Leu Lys Asp Ala Glu 610 615 620 Lys Cys Ile
 Glu Leu Lys Pro Asp Phe Val Lys Gly Tyr Ala Arg Lys 625 630 635 640 Gly His Ala Tyr
 Phe Trp Thr Lys Gln Tyr Asn Arg Ala Leu Gln Ala 645 650 655 Tyr Asn Glu Gly Leu Lys
 Val Asp Pro Ser Asn Ala Asp Cys Lys Asp 660 665 670 Gly Arg Tyr Arg Thr Ile Met Lys
 Ile Gln Glu Met Ala Ser Gly Gln 675 680 685 Ser Ala Asp Gly Asp Glu Ala Ala Arg Arg
 Ala Met Asp Asp Pro Glu 690 695 700 Ile Ala Ala Ile Met Gln Asp Ser Tyr Met Gln Leu
 Val Leu Lys Glu 705 710 715 720 Met Gln Asn Asp Pro Thr Arg Ile Gln Glu Tyr Met Lys
 Asp Ser Gly 725 730 735 Ile Ser Ser Lys Ile Asn Lys Leu Ile Ser Ala Gly Ile Ile Arg
 Phe 740 745 750 Gly Gln Glu Phe Ser Leu Thr Asp Pro Ala Val Leu Gly Glu Glu Thr 755
 760 765 His Leu Arg Val Arg Val Val Pro Asp Lys Ala Asn Lys Thr Leu Thr 770 775 780
 Val Glu Asp Asn Gly Ile Gly Met Thr Lys Ala Asp Leu Val Asn Asn 785 790 795 800 Leu
 Gly Thr Ile Ala Arg Ser Gly Thr Lys Ala Phe Met Glu Ala Leu 805 810 815 Glu Ala Gly
 Gly Asp Met Ser Met Ile Gly Gln Phe Gly Val Gly Phe 820 825 830 Tyr Ser Ala Tyr Leu
 Val Ala Asp Arg Val Thr Val Val Ser Lys Asn 835 840 845 Asn Ser Asp Glu Ala Tyr Val
 Trp Glu Ser Ser Ala Gly Gly Thr Phe 850 855 860 Thr Ile Thr Ser Val Pro Glu Ser Asp
 Met Lys Arg Gly Thr Arg Ile 865 870 875 880 Thr Leu His Leu Lys Glu Asp Gln Gln Glu
 Tyr Leu Glu Glu Arg Arg 885 890 895 Val Lys Glu Leu Ile Lys Lys His Ser Glu Phe Ile
 Gly Tyr Asp Ile 900 905 910 Glu Leu Met Val Glu Lys Thr Ala Glu Lys Glu Val Thr Asp
 Glu Asp 915 920 925 Glu Glu Glu Asp Glu Ser Lys Lys Lys Ser Cys Gly Asp Glu Gly Glu
 930 935 940 Pro Lys Val Glu Glu Val Thr Glu Gly Gly Glu Asp Lys Lys Lys Lys 945 950
 955 960 Thr Lys Lys Val Lys Glu Val Thr Lys Thr Tyr Glu Val Gln Asn Lys 965 970 975
 His Lys Pro Leu Trp Thr Arg Asp Pro Lys Asp Val Thr Lys Glu Glu 980 985 990 Tyr Ala

Ala Phe Tyr Lys Ala Ile Ser Asn Asp Trp Glu Asp Pro Ala 995 1000 1005 Ala Thr Lys
 His Phe Ser Val Glu Gly Gln Leu Glu Phe Arg Ala Ile 1010 1015 1020 Ala Phe Val Pro
 Lys Arg Ala Pro Phe Asp Met Phe Glu Pro Asn Lys 1025 1030 1035 1040 Lys Arg Asn Asn
 Ile Lys Leu Tyr Val Arg Arg Val Phe Ile Met Asp 1045 1050 1055 Asn Cys Glu Asp Leu
 Cys Pro Asp Trp Leu Gly Phe Val Lys Gly Val 1060 1065 1070 Val Asp Ser Glu Asp Leu
 Pro Leu Asn Ile Ser Arg Glu Asn Leu Gln 1075 1080 1085 Gln Asn Lys Ile Leu Lys Val
 Ile Arg Lys Asn Ile Val Lys Lys Cys 1090 1095 1100 Leu Glu Leu Phe Glu Glu Ile Ala
 Glu Asn Lys Glu Asp Tyr Lys Gln 1105 1110 1115 1120 Phe Tyr Glu Gln Phe Gly Lys Asn
 Ile Lys Leu Gly Ile His Glu Asp 1125 1130 1135 Thr Ala Asn Arg Lys Lys Leu Met Glu
 Leu Leu Arg Phe Tyr Ser Thr 1140 1145 1150 Glu Ser Gly Glu Glu Met Thr Thr Leu Lys
 Asp Tyr Val Thr Arg Met 1155 1160 1165 Lys Pro Glu Gln Lys Ser Ile Tyr Tyr Ile Thr
 Gly Asp Ser Lys Lys 1170 1175 1180 Lys Leu Glu Ser Ser Pro Phe Ile Glu Lys Ala Arg
 Arg Cys Gly Leu 1185 1190 1195 1200 Glu Val Leu Phe Met Thr Glu Pro Ile Asp Glu Tyr
 Val Met Gln Gln 1205 1210 1215 Val Lys Asp Phe Glu Asp Lys Lys Phe Ala Cys Leu Thr
 Lys Glu Gly 1220 1225 1230 Val His Phe Glu Glu Ser Glu Glu Glu Lys Lys Gln Arg Glu
 Glu Lys 1235 1240 1245 Lys Ala Ala Cys Glu Lys Leu Cys Lys Thr Met Lys Glu Val Leu
 Gly 1250 1255 1260 Asp Lys Val Glu Lys Val Thr Val Ser Glu Arg Leu Ser Thr Ser Pro
 1265 1270 1275 1280 Cys Ile Leu Val Thr Ser Glu Phe Gly Trp Ser Ala His Met Glu Gln
 1285 1290 1295 Ile Met Arg Asn Gln Ala Leu Arg Asp Ser Ser Met Ala Gln Tyr Met 1300
 1305 1310 Val Ser Lys Lys Thr Met Glu Val Asn Pro Asp His Pro Ile Ile Lys 1315 1320
 1325 Glu Leu Arg Arg Val Glu Ala Asp Glu Asn Asp Lys Ala Val Lys 1330 1335 1340
 Asp Leu Val Phe Leu Leu Phe Asp Thr Ser Ser Leu Leu Thr Ser Gly Phe 1345 1350 1355 1360
 Gln Leu Asp Asp Pro Thr Gly Tyr Ala Glu Arg Ile Asn Arg Met Ile 1365 1370 1375 Lys
 Leu Gly Leu Ser Leu Asp Glu Glu Glu Glu Glu Val Ala Glu Ala 1380 1385 1390 Pro Pro
 Ala Glu Ala Ala Pro Ala Glu Val Thr Ala Gly Thr Ser Ser 1395 1400 1405 Met Glu Gln
 Val Asp Asp Ile Met Ala Gln Asn Asp Lys Ile Ala Pro 1410 1415 1420 Gln Asp Gln Asp
 Ser Phe Leu Asp Asp Gln Pro Gly Val Arg Pro Ile 1425 1430 1435 1440 Pro Ser Phe Asp
 Asp Met Pro Leu His Gln Asn Leu Leu Arg Gly Ile 1445 1450 1455 Tyr Ser Tyr Gly Phe
 Glu Lys Pro Ser Ser Ile Gln Gln Arg Ala Ile 1460 1465 1470 Ala Pro Phe Thr Arg Gly
 Gly Asp Ile Ile Ala Gln Ala Gln Ser Gly 1475 1480 1485 Thr Gly Lys Thr Gly Ala Phe
 Ser Ile Gly Leu Leu Gln Arg Leu Asp 1490 1495 1500 Phe Arg His Asn Leu Ile Gln Gly
 Leu Val Leu Ser Pro Thr Arg Glu 1505 1510 1515 1520 Leu Ala Leu Gln Thr Ala Glu Val
 Ile Ser Arg Ile Gly Glu Phe Leu 1525 1530 1535 Ser Asn Ser Ser Lys Phe Cys Glu Thr
 Phe Val Gly Gly Thr Arg Val 1540 1545 1550 Gln Asp Asp Leu Arg Lys Leu Gln Ala Gly
 Val Ile Val Ala Val Gly 1555 1560 1565 Thr Pro Gly Arg Val Ser Asp Val Ile Lys Arg
 Gly Ala Leu Arg Thr 1570 1575 1580 Glu Ser Leu Arg Val Leu Val Leu Asp Glu Ala Asp
 Glu Met Leu Ser 1585 1590 1595 1600 Gln Gly Phe Ala Asp Gln Ile Tyr Glu Ile Phe Arg
 Phe Leu Pro Lys 1605 1610 1615 Asp Ile Gln Val Ala Leu Phe Ser Ala Thr Met Pro Glu
 Glu Val Leu 1620 1625 1630 Glu Leu Thr Lys Lys Phe Met Arg Asp 1635 1640 <200>
 SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 97 <211> LENGTH: 1427 <212> TYPE: PRT
 <213> ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: Fusion
 (poly-protein) constructs comprising multiple Leishmania antigens <400> SEQUENCE:
 97 Met His His His His His His Met Ser Cys Gly Asn Ala Lys Ile Asn 5 10 15 Ser Pro
 Ala Pro Ser Phe Glu Glu Val Ala Leu Met Pro Asn Gly Ser 20 25 30 Phe Lys Lys Ile
 Ser Leu Ser Ser Tyr Lys Gly Lys Trp Val Val Leu 35 40 45 Phe Phe Tyr Pro Leu Asp
 Phe Thr Phe Val Cys Pro Thr Glu Val Ile 50 55 60 Ala Phe Ser Asp Ser Val Ser Arg
 Phe Asn Glu Leu Asn Cys Glu Val 65 70 75 80 Leu Ala Cys Ser Ile Asp Ser Glu Tyr Ala
 His Leu Gln Trp Thr Leu 85 90 95 Gln Asp Arg Lys Lys Gly Gly Leu Gly Thr Met Ala
 Ile Pro Met Leu 100 105 110 Ala Asp Lys Thr Lys Ser Ile Ala Arg Ser Tyr Gly Val Leu
 Glu Glu 115 120 125 Ser Gln Gly Val Ala Tyr Arg Gly Leu Phe Ile Ile Asp Pro His Gly
 130 135 140 Met Leu Arg Gln Ile Thr Val Asn Asp Met Pro Val Gly Arg Ser Val 145 150
 155 160 Glu Glu Val Leu Arg Leu Leu Glu Ala Phe Gln Phe Val Glu Lys His 165 170 175
 Gly Glu Val Cys Pro Ala Asn Trp Lys Lys Gly Ala Pro Thr Met Lys 180 185 190 Pro Glu
 Pro Asn Ala Ser Val Glu Gly Tyr Phe Ser Lys Gln Gly Ser 195 200 205 Met Asp Ala Thr
 Glu Leu Lys Asn Lys Gly Asn Glu Glu Phe Ser Ala 210 215 220 Gly Arg Tyr Val Glu Ala
 Val Asn Tyr Phe Ser Lys Ala Ile Gln Leu 225 230 235 240 Asp Glu Gln Asn Ser Val Leu
 Tyr Ser Asn Arg Ser Ala Cys Phe Ala 245 250 255 Ala Met Gln Lys Tyr Lys Asp Ala Leu
 Asp Asp Ala Asp Lys Cys Ile 260 265 270 Ser Ile Lys Pro Asn Trp Ala Lys Gly Tyr Val
 Arg Arg Gly Ala Ala 275 280 285 Leu His Gly Met Arg Arg Tyr Asp Asp Ala Ile Ala Ala
 Tyr Glu Lys 290 295 300 Gly Leu Lys Val Asp Pro Ser Asn Ser Gly Cys Ala Gln Gly Val
 Lys

Detailed Description Paragraph Table (15):

305 310 315 320 Asp Val Gln Val Ala Lys Ala Arg Glu Ala Arg Asp Pro Ile Ala Arg 325

h e b b g e e e f c e

e ge

330 335 Val Phe Thr Pro Glu Ala Phe Arg Lys Ile Gln Glu Asn Pro Lys Leu 340 345 350
Ser Leu Leu Met Leu Gln Pro Asp Tyr Val Lys Met Val Asp Thr Val 355 360 365 Ile Arg
Asp Pro Ser Gln Gly Arg Leu Tyr Met Glu Asp Gln Arg Phe 370 375 380 Ala Leu Thr Leu
Met Tyr Leu Ser Gly Met Lys Ile Pro Asn Asp Gly 385 390 395 400 Asp Gly Glu Glu Glu
Glu Arg Pro Ser Ala Lys Ala Ala Glu Thr Ala 405 410 415 Lys Pro Lys Glu Glu Lys Pro
Leu Thr Asp Asn Glu Lys Glu Ala Leu 420 425 430 Ala Leu Lys Glu Glu Gly Asn Lys Leu
Tyr Leu Ser Lys Lys Phe Glu 435 440 445 Glu Ala Leu Thr Lys Tyr Gln Glu Ala Gln Val
Lys Asp Pro Asn Asn 450 455 460 Thr Leu Tyr Ile Leu Asn Val Ser Ala Val Tyr Phe Glu
Gln Gly Asp 465 470 475 480 Tyr Asp Lys Cys Ile Ala Glu Cys Glu His Gly Ile Glu His
Gly Arg 485 490 495 Glu Asn His Cys Asp Tyr Thr Ile Ile Ala Lys Leu Met Thr Arg Asn
500 505 510 Ala Leu Cys Leu Gln Arg Gln Arg Lys Tyr Glu Ala Ala Ile Asp Leu 515 520
525 Tyr Lys Arg Ala Leu Val Glu Trp Arg Asn Pro Asp Thr Leu Lys Lys 530 535 540 Leu
Thr Glu Cys Glu Lys Glu His Gln Lys Ala Val Glu Glu Ala Tyr 545 550 555 560 Ile Asp
Pro Glu Ile Ala Lys Gln Lys Lys Asp Glu Gly Asn Gln Tyr 565 570 575 Phe Lys Glu Asp
Lys Phe Pro Glu Ala Val Ala Ala Tyr Thr Glu Ala 580 585 590 Ile Lys Arg Asn Pro Ala
Glu His Thr Ser Tyr Ser Asn Arg Ala Ala 595 600 605 Ala Tyr Ile Lys Leu Gly Ala Phe
Asn Asp Ala Leu Lys Asp Ala Glu 610 615 620 Lys Cys Ile Glu Leu Lys Pro Asp Phe Val
Lys Gly Tyr Ala Arg Lys 625 630 635 640 Gly His Ala Tyr Phe Trp Thr Lys Gln Tyr Asn
Arg Ala Leu Gln Ala 645 650 655 Tyr Asn Glu Gly Leu Lys Val Asp Pro Ser Asn Ala Asp
Cys Lys Asp 660 665 670 Gly Arg Tyr Arg Thr Ile Met Lys Ile Gln Glu Met Ala Ser Gly
Gln 675 680 685 Ser Ala Asp Gly Asp Glu Ala Ala Arg Arg Ala Met Asp Asp Pro Glu 690
695 700 Ile Ala Ala Ile Met Gln Asp Ser Tyr Met Gln Leu Val Leu Lys Glu 705 710 715
720 Met Gln Asn Asp Pro Thr Arg Ile Gln Glu Tyr Met Lys Asp Ser Gly 725 730 735 Ile
Ser Ser Lys Ile Asn Lys Leu Ile Ser Ala Gly Ile Ile Arg Phe 740 745 750 Gly Gln Glu
Phe Ser Leu Thr Asp Pro Ala Val Leu Gly Glu Glu Thr 755 760 765 His Leu Arg Val Arg
Val Val Pro Asp Lys Ala Asn Lys Thr Leu Thr 770 775 780 Val Glu Asp Asn Gly Ile Gly
Met Thr Lys Ala Asp Leu Val Asn Asn 785 790 795 800 Leu Gly Thr Ile Ala Arg Ser Gly
Thr Lys Ala Phe Met Glu Ala Leu 805 810 815 Glu Ala Gly Gly Asp Met Ser Met Ile Gly
Gln Phe Gly Val Gly Phe 820 825 830 Tyr Ser Ala Tyr Leu Val Ala Asp Arg Val Thr Val
Val Ser Lys Asn 835 840 845 Asn Ser Asp Glu Ala Tyr Val Trp Glu Ser Ser Ala Gly Gly
Thr Phe 850 855 860 Thr Ile Thr Ser Val Pro Glu Ser Asp Met Lys Arg Gly Thr Arg Ile
865 870 875 880 Thr Leu His Leu Lys Glu Asp Gln Gln Glu Tyr Leu Glu Glu Arg Arg 885
890 895 Val Lys Glu Leu Ile Lys Lys His Ser Glu Phe Ile Gly Tyr Asp Ile 900 905 910
Glu Leu Met Val Glu Lys Thr Ala Glu Lys Glu Val Thr Asp Glu Asp 915 920 925 Glu Glu
Glu Asp Glu Ser Lys Lys Lys Ser Cys Gly Asp Glu Gly Glu 930 935 940 Pro Lys Val Glu
Glu Val Thr Glu Gly Gly Glu Asp Lys Lys Lys Lys 945 950 955 960 Thr Lys Lys Val Lys
Glu Val Thr Lys Thr Tyr Glu Val Gln Asn Lys 965 970 975 His Lys Pro Leu Trp Thr Arg
Asp Pro Lys Asp Val Thr Lys Glu Glu 980 985 990 Tyr Ala Ala Phe Tyr Lys Ala Ile Ser
Asn Asp Trp Glu Asp Pro Ala 995 1000 1005 Ala Thr Lys His Phe Ser Val Glu Gly Gln
Leu Glu Phe Arg Ala Ile 1010 1015 1020 Ala Phe Val Pro Lys Arg Ala Pro Phe Asp Met
Phe Glu Pro Asn Lys 1025 1030 1035 1040 Lys Arg Asn Asn Ile Lys Leu Tyr Val Arg Arg
Val Phe Ile Met Asp 1045 1050 1055 Asn Cys Glu Asp Leu Cys Pro Asp Trp Leu Gly Phe
Val Lys Gly Val 1060 1065 1070 Val Asp Ser Glu Asp Leu Pro Leu Asn Ile Ser Arg Glu
Asn Leu Gln 1075 1080 1085 Gln Asn Lys Ile Leu Lys Val Ile Arg Lys Asn Ile Val Lys
Lys Cys 1090 1095 1100 Leu Glu Leu Phe Glu Glu Ile Ala Glu Asn Lys Glu Asp Tyr Lys
Gln 1105 1110 1115 1120 Phe Tyr Glu Gln Phe Gly Lys Asn Ile Lys Leu Gly Ile His Glu
Asp 1125 1130 1135 Thr Ala Asn Arg Lys Lys Leu Met Glu Leu Leu Arg Phe Tyr Ser Thr
1140 1145 1150 Glu Ser Gly Glu Glu Met Thr Thr Leu Lys Asp Tyr Val Thr Arg Met 1155
1160 1165 Lys Pro Glu Gln Lys Ser Ile Tyr Tyr Ile Thr Gly Asp Ser Lys Lys 1170 1175
1180 Lys Leu Glu Ser Ser Pro Phe Ile Glu Lys Ala Arg Arg Cys Gly Leu 1185 1190 1195
1200 Glu Val Leu Phe Met Thr Glu Pro Ile Asp Glu Tyr Val Met Gln Gln 1205 1210 1215
Val Lys Asp Phe Glu Asp Lys Lys Phe Ala Cys Leu Thr Lys Glu Gly 1220 1225 1230 Val
His Phe Glu Glu Ser Glu Glu Glu Lys Lys Gln Arg Glu Glu Lys 1235 1240 1245 Lys Ala
Ala Cys Glu Lys Leu Cys Lys Thr Met Lys Glu Val Leu Gly 1250 1255 1260 Asp Lys Val
Glu Lys Val Thr Val Ser Glu Arg Leu Ser Thr Ser Pro 1265 1270 1275 1280 Cys Ile Leu
Val Thr Ser Glu Phe Gly Trp Ser Ala His Met Glu Gln 1285 1290 1295 Ile Met Arg Asn
Gln Ala Leu Arg Asp Ser Ser Met Ala Gln Tyr Met 1300 1305 1310 Val Ser Lys Lys Thr
Met Glu Val Asn Pro Asp His Pro Ile Ile Lys 1315 1320 1325 Glu Leu Arg Arg Val
Glu Ala Asp Glu Asn Asp Lys Ala Val Lys 1330 1335 1340 Asp Leu Val Phe Leu Leu Phe
Asp Thr Ser Leu Leu Thr Ser Gly Phe 1345 1350 1355 1360 Gln Leu Asp Asp Pro Thr Gly
Tyr Ala Glu Arg Ile Asn Arg Met Ile 1365 1370 1375 Lys Leu Gly Leu Ser Leu Asp Glu
Glu Glu Glu Glu Val Ala Glu Ala 1380 1385 1390 Pro Pro Ala Glu Ala Ala Pro Ala Glu
Val Thr Ala Gly Thr Ser Ser 1395 1400 1405 Met Glu Gln Val Asp Asp Ile His His Thr
Gly Gly Arg Ser Ser Arg 1410 1415 1420 Ser Gly Cys 1425 <200> SEQUENCE

CHARACTERISTICS: <210> SEQ ID NO 98 <211> LENGTH: 4929 <212> TYPE: DNA <213>

ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: DNA sequence encoding fusion (poly-protein) constructs comprising multiple Leishmania antigens

<400> SEQUENCE: 98 catatgcacc accaccacca ccacatgtcc tgcggtaacg ccaagatcaa
ctctcccgcg 60 ccgtccttcg aggaggtggc gtcctatgcc aacggcagct tcaagaagat cagcctctcc 120
tcctacaagg gcaagtgggt cgtgctcttc ttctacccgc tcgacttcac cttcgtgtgc 180 ccgacagagg
tcctcgcgtt ctccgacagc gtgagtcgct tcaacgagct caactgcgag 240 gtcctcgcgt gtcgataga
cagcgagtac gcgcacctgc agtggacgct gcaggaccgc 300 aagaagggcg gcctcgggac catggcgatc
ccaatgctag ccgacaagac caagagcatc 360 gtcgtttcct acggcgtgct ggaggagagc cagggcggtg
cctaccgcgg tctcttcac 420 atcgaccccc atggcatgct gcgtcagatc accgtcaatg acatgccggt
gggcccgcagc 480 gtggaggagg ttctacgcct gctggaggct tttcagttcg tggagaagca cggcgagggtg
540 tgccccgcga actggaagaa gggcgcccc acgatgaagc cggaaccgaa tgcgtctgtc 600
gagggatact tcagcaagca gggatccatg gacgcaactg agctgaagaa caaggggaac 660 gaagagttct
ccgccggccg ctatgtggag gcgtgtaact acttctcaaa ggcatccag 720 ttggatgagc agaacagtgt
cctctacagc aaccgctccg cctgttttgc agcatgcag 780 aaatacaagg acgctgtgga cgacgccgac
aagtgcattc cgatcaagcc gaattggggc 840 aagggctacg tgcgccgagg agcagctctc catggcatgc
gccgtacga cgatgccatt 900 gccgctatg aaaaggggct caaggtggac cttccaaca gcggctgcgc
gcagggcgtg 960 aaggacgtgc aggtagccaa ggcccgcgaa gcacgtgacc ccacgctcgc cgtcttcacc
1020 ccggaggcgt tccgcaagat ccaagagaat cccaagctgt ctctacttat gctgcagccg 1080
gactacgtga agatggtaga caccgtcatc cgcgaccctt cgcagggccg gctgtacatg 1140 gaagaccagc
gctttgcctt gacgtcatg tacctgagcg gaatgaagat tcccaacgat 1200 ggtgatggcg aggaggagga
acgtccgtct gcgaaggcgg cagagacagc gaagccaaaa 1260 gaggagaagc ctctcaccga caacgagaag
gagggccctg gcgtcaagga ggagggcaac 1320 aagctgtacc tctcgaagaa gtttgaggag gcgctgacca
agtaccaaga ggcgaggtg 1380 aaagacccca acaacacttt atacattctg aacgtgtcgg ccgtgtactt
cgagcagggt 1440 gactacgaca agtgcctcgc cgagtgcgag cacggtatcg agcaggtcgc cgagaaccac
1500 tgcgactaca caatcattgc gaagctcatg acccggaacg ccttgtgcct ccagaggtcag 1560
aggaagtacg aggtgctat cgacctttac aagcgcgccc ttgtcgagtg gcgtaaccct 1620 gacacctca
agaagctgac ggagtgcgag aaggagcacc aaaagggcgt ggaggaagcc 1680 tacatcgatc ctgagatcgc
gaagcagaag aaagacgaag gtaaccagta cttcaaggag 1740 gataagttcc ccgaggccgt ggcagcgta
acggaggcca tcaagcgcaa ccctgccgag 1800 cacacctcct acagcaatcg cgcggccgcg tacatcaagc
ttggagcctt caacgacgcc 1860 ctcaaggacg cggagaagtg cattgagctg aagcccgact ttgttaaggg
ctacgcgcgc 1920 aagggtcatg ctactttttg gaccaagcag tacaaccgcg cgctgcaggc gtacaatgag
1980 ggcctcaagg tggaccgag caatgcggac tgcaaggatg ggcggtatcg cacaatcatg 2040
aagattcagg agatggcatc tggccaatcc gcggtggcg acgagggcgc gcgccgggccc 2100 atggacgatc
ctgaaatcgc ggcaatcatg caagatagct acatgcaact agtggtgaag 2160 gagatgcaga acgatccac
gcgcattcag gactacatga aggactccg gatctcatcg 2220 aagatcaaca agctgatttc agctggcatc
attcgttttg gtcaggaatt cagcctgacg 2280 gacccggcgg tgctgggcca ggagactcac ctgcgcgtcc
gcgtggtgcc ggacaaggcg 2340 aacaagacgc tgacggtgga ggataacggc atcggcagta ccaaggcgga
cctcgtgaac 2400 aatctgggca cgatcgcgcg ctccggcacg aaggcggttca tggaggcact ggaggccggc
2460 ggcgacatga gcatgatcgg ccagttcgtt gtcggcttct actccgcgta ccttgtggcg 2520
gaccgcgtga cggtggtgct gaagaacaac tcggacgagg cgtacgtatg ggagtcgtcc 2580 gcgggcggca
cgttccat caagagctg cgagagtcgg acatgaagcg cggcacgcgc 2640 atcacgtgc acctaaagga
ggaccagcag gactacctgg aggagcggcg ggtgaaggag 2700 ctgatcaaga agcatccga gttcatcggc
tacgacatcg agctgatggt ggagaagacg 2760 gcggagaagg aggtgacgga cgaggacgag gaggaggagc
agtcgaagaa gaagtcctgc 2820 ggggacgagg gcgagccgaa ggtggaggag gtgacggagg gcggcgagga
caagaagaag 2880 aagacgaaga aggtgaagga ggtgacgaag acgtacgagg tccagaacaa gcacaagccg
2940 ctctggacgc gcgacccgaa ggacgtgacg aaggaggagt acgcggcctt ctacaaggcc 3000
atctccaacg actgggagga cccggcgggc acgaagcact tctcgttgga gggccagctg 3060 gaggttccgcg
cgatcgctt cgtgccgaag cgcgcgccgt tcgacatggt cgagccgaac 3120 aagaagcgca acaacatcaa
gctgtacgtg cgccgcgtgt tcatcatgga caactgcgag 3180 gacctgtgcc cggactggct cggcttcgtg
aagggcgctg tggacagcga ggacctgccg 3240 ctgaacatct cgcgcgagaa cctgcagcag aacaagatcc
tgaaggatgat ccgcaagaac 3300 atcgtgaaga agtgcttggg gctgttcgaa gagatagcgg agaacaagga
ggactacaag 3360 cagttctacg agcagttcgg caagaacatc aagctgggca tccacgagga cacggcgaac
3420 cgcaagaagc tgatggagtt gctgcgcttc tacagcaccg agtcggggga ggagatgacg 3480
acactgaagg actacgtgac gcgcagtaag ccggagcaga agtcgatcta ctacatcact 3540 ggcgacagca
agaagaagct ggagtcgtcg ccgttcacg agaaggcgag acgctgcggg 3600 ctcgaggtgc tgttcagac
ggagccgatc gacgagtacg tgatgcagca ggtgaaggac 3660 ttcgaggaca agaagttcgc gtgcctgacg
aaggaaggcg tgcacttcga ggagtcggag 3720 gaggagaaga agcagcgca ggagaagaag gcggcggtgc
agaagctgtg caagacgatg 3780 aaggaggtgc tgggcgacaa ggtggagaag gtgaccgtgt cggagcgct
gtcgacgtcg 3840 ccgtgcatcc tggtagctgc ggagtttggg tggtcggcg acatggaaca gatcgcgc
3900 aaccaggcgc tgcgcgactc cagcatggcg cagtacatgg tgtccaagaa gacgatggag 3960
gtgaaccccg accaccccat catcaaggag ctgcgccgcc gcgtggaggc ggacgagaac 4020 gacaaggccg
tgaaggacct cgtcttcctg ctcttcgaca cgctcgtgct cagctccggc 4080 ttccagctgg atgacccac
cggctacgcc gagcgcatca accgcatgat caagctcggc 4140 ctgtcgtcgc acgaggagga ggaggaggtc

gccgaggcgc cgccggccga ggcagcccc 4200 gcggagggtca ccgcccggcac ctccagcatg gagcagggtg
 acgatatcat ggcgcagaat 4260 gataagatcg cccccagga ccaggactcc ttcctcgatg accagcccgg
 cgttcgcccc 4320 atcccgctct tcgacgacat ccgctgcac cagaacctgc tgcgtggcat ctactcgta
 4380 gggttcgaga agccgtccag catccagcag cgcgcgatag ccccttcac gcgcggcggc 4440
 gacatcatcg cgcaggccca gtccgggtacc ggcaagacgg gtgccttctc catcggtctg 4500 ctgcagcgcc
 tggacttccg ccacaacctg atccaggggc tegtgtctc cccactcgc 4560 gagctggccc tgcagacggc
 ggaggtgatc agccgcacg gtgagttct gtcgaacagc 4620 tccaagttct gcgagacct tgtcggcggc
 acgcgcgtgc aggatgacct gcgcaagctg 4680 caggccggcg tcatcggtgc cgtgggcacg ccggggccgcg
 tgtccgacgt gatcaagcgt 4740 ggcgcgtgc gcacagagtc gctgcgcgtg ctggtgctcg acgaggctga
 tgagatgctg 4800 tctcagggtc tcgcgacca gatttacgag atcttccgct tctgcccga ggacatccag
 4860 gtcgcgtct tctccgccac gatgccggag gaggtactgg agctgacgaa gaagttcatg 4920
 cgcgactaa 4929 <200> SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 99 <211> LENGTH:
 4233 <212> TYPE: DNA <213> ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER
 INFORMATION: DNA sequence encoding fusion (poly-protein) constructs comprising
 multiple Leishmania antigens <400> SEQUENCE: 99 ccagtgtggt ggatgtcctg cggtaacgcc
 aagatcaact ctcccgcgcc gtccttcgag 60 gaggtggcgc tcatgcccga cggcagcttc aagaagatca
 gcctctctc ctacaagggc 120 aagtgggtcg tgccttctt ctaccgcgtc gacttcacct tegtgtgccc
 gacagaggtc 180 atcgcttct ccgacagcgt gctgcgttc aacgagctca actgcgaggt cctcgcgtgc
 240 tcgatagaca gcgagtacgc gcacctgcag tggacgtgc aggaccgcaa gaaggcggc 300
 ctggggacca tgggatccc aatgctagcc gacaagacca agagcatcgc tcgttcctac 360 ggctgtctgg
 aggagagcca gggcgtggc taccgcgtc tcttcatcat cgacccccat 420 ggcagctgc gtcagatcac
 cgtcaatgac atgccgttg gccgcagcgt ggaggaggtt 480 ctacgcctgc tggaggcttt tcagttcgtg
 gagaagcacg gcgaggtgtg cccgcgaac 540

Detailed Description Paragraph Table (16):

tggaagaagg gcgccccac gatgaagcgg gaaccgaatg cgtctgtcga gggatacttc 600 agcaagcagg
 gatccatgga cgcaactgag ctgaagaaca aggggaacga agagttctcc 660 gccggccgct atgtggaggc
 ggtgaactac ttctcaaagg cgatccagtt ggatgagcag 720 aacagtgtcc tctacagcaa ccgctccgcc
 tgttttgag ccatgcagaa atacaaggac 780 gcgctggacg acgcccagaa gtgcatctcg atcaagccga
 attgggcaa gggctacgtg 840 cgccgaggag cagctctcca tggcatgcgc cgctacgacg atgccattgc
 cgcgtatgaa 900 aaggggtca aggtggacc ttccaacagc ggctgcgcgc agggcgtgaa ggacgtgcag
 960 gtagccaagg cccgcgaagc acgtgacccc atcgctcgcg tcttcacccc ggaggcgttc 1020
 cgcaagatcc aagagaatcc caagctgtct ctacttatgc tgcagccgga ctacgtgaag 1080 atggtagaca
 ccgtcatccg cgacccttcg cagggccggc tgtacatgga agaccagcgc 1140 tttgccctga cgctcatgta
 cctgagcgga atgaagattc ccaacgatgg tgatggcgag 1200 gaggaggaac gtccgtctgc gaaggcggca
 gagacagcga agccaaaaga ggagaagcct 1260 ctaccgcaca acgagaagga ggccctggcg ctcaaggagg
 agggcaacaa gctgtacctc 1320 tcgaagaagt ttgaggaggc gctgaccaag taccaagagg cgcagggtgaa
 agacccaac 1380 aacactttat acattctgaa cgtgtcggcc gtgtacttcg agcagggtga ctacgacaag
 1440 tgcctgcgg agtgcgagca cggtatcgag cacggtcgcg agaaccactg cgactacaca 1500
 atcattgca agctcatgac ccggaacgcg ttgtgcctcc agaggcagag gaagtacgag 1560 gctgctatcg
 acctttaca gcgcgccctt gtcgagtgcc gtaacctga caccctcaag 1620 aagctgacgg agtgcgagaa
 ggagcaccaa aaggcgttg aggaagccta catcgatcct 1680 gagatcgca agcagaagaa agacgaaggt
 aaccagtact tcaaggagga taagttcccc 1740 gaggccgtgg cagcgtacac ggaggccatc aagcgcaacc
 ctgccgagca cacctctac 1800 agcaatcgcg cgcccgcgta catcaagctt ggagccttca acgacgccct
 caaggacgcg 1860 gagaagtgca ttgagctgaa gcccgaactt gttaagggtt acgcgcgcaa gggtcagtct
 1920 tacttttgga ccaagcagta caaccgcgcg ctgcaggcgt acaatgaggg cctcaagggtg 1980
 gaccgagca atgcggactg caaggatggg cggtatcgca caatcatgaa gattcaggag 2040 atggcatctg
 gccaatccgc ggatggcgac gaggcggcgc gccgggccat ggacgatcct 2100 gaaatcgcg caatcatgca
 agatagctac atgcaactag tgttgaagga gatgcagaac 2160 gatccacgc gcattcagga gtacatgaag
 gactccggga tctcatcgaa gatcaacaag 2220 ctgatttcag ctggcatcat tcgttttggt cagggaattca
 gcctgacgga cccggcgggtg 2280 ctgggcgagg agactcacct gcgcgtccgc gtggtgccgg acaaggcgaa
 caagacgctg 2340 acggtggagg ataacggcat cggcagatgacc aaggcggacc tcgtgaacaa tctgggcacg
 2400 atcgcgcgct ccggcacgaa ggcgttcatt gaggcactgg aggcggcgcg cgacatgagc 2460
 atgatcggcc agttcggtgt cggttctac tccgcgtacc ttgtggcgga ccgcgtgacg 2520 gtggtgtcga
 agaacaactc ggacgaggcg tacgtatggg agtcgtccgc gggcggcacg 2580 ttcaccatca cgagcgtgcc
 ggagtcggac atgaagcgcg gcacgcgat cacgtgcac 2640 ctaaaggagg accagcagga gtacctggag
 gagcgcggg tgaaggagct gatcaagaag 2700 cactccgagt tcatcggtta cgacatcgag ctgatggtg
 agaagacggc ggagaaggag 2760 gtgacggagc aggcagga ggaggacgag tcgaagaaga agtctgcgg
 ggacgagggc 2820 gagccgaagg tggaggagt cagcggggc ggcgaggaca agaagaaga gacgaagaag
 2880 gtgaaggagg tgacgaagac gtacgaggtc cagaacaagc acaagccgct ctggagcgc 2940
 gaccgaagg acgtgacgaa ggaggagtac gcggccttct acaaggccat ctccaacgac 3000 tgggaggacc
 cggcggcgac gaagcacttc tcggtggagg gccagctgga gttccgcgcg 3060 atcgcgcttc tgccgaagcg
 cgcgcgcttc gacatgttc agccgaacaa gaagcgcaac 3120 aacatcaagc tgtacgtgcg ccgcgtgttc
 atcatggaca actgcgagga cctgtgccc 3180 gactggctcg gttcgtgaa gggcgtcgtg gacagcgagg

acctgccgct gaacatctcg 3240 cgcgagaacc tgcagcagaa caagatcctg aaggtgatcc gcaagaacat
cgtgaagaag 3300 tgcctggagc tgctcgaaga gatagcggag aacaaggagg actacaagca gttctacgag
3360 cagttcggca agaacatcaa gctgggcac cagcaggaca cggcgaaccg caagaagctg 3420
atggaggtgc tgcgcttcta cagcaccgag tcgggggagg agatgacgac actgaaggac 3480 tacgtgacgc
gcatgaagcc ggagcagaag tcgatctact acatcactgg cgacagcaag 3540 aagaagctgg agtcgtcgcc
gttcacgag aaggcgagac gctgcgggct cgaggtgctg 3600 ttcacgacgg agccgatcga cgagtacgtg
atgcagcagg tgaaggactt cgaggacaag 3660 aagttcgcgt gcctgacgaa ggaaggcgtg cacttcgagg
agtccgagga ggagaagaag 3720 cagcgcgagg agaagaaggc ggcgtgagag aagctgtgca agacgatgaa
ggaggtgctg 3780 ggcgacaagg tggagaaggt gaccgtgtcg gagcgctgt cgacgtcgcc gtgcatcctg
3840 gtgacgtcgg agtttgggtg gtcgggcgac atggaacaga tcatgacgaa ccaggcgctg 3900
cgcgactcca gcatggcgca gtacatggtg tccaagaaga cgatggaggt gaaccccgac 3960 caccatca
tcaaggagct gcgcgcgcgt gtggaggcgg acgagaacga caaggccgtg 4020 aaggacctcg tcttctgtg
cttcgacacg tcgctgctca cgtccggctt ccagctggat 4080 gacccaccg gctacgccga gcgcatcaac
cgcatgatca agtcggcct gtcgctcgac 4140 gaggaggagg aggaggtcgc cgaggcgccg ccggccgagg
cagccccgc ggaggtcacc 4200 gccggcacct ccagcatgga gcaggtggac taa 4233 <200> SEQUENCE
CHARACTERISTICS: <210> SEQ ID NO 100 <211> LENGTH: 4917 <212> TYPE: DNA <213>
ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: DNA sequence
encoding fusion (poly-protein) constructs comprising multiple Leishmania antigens
<400> SEQUENCE: 100 ccagtgtggt ggatgtcctg cgtaacgcc aagatcaact ctcccgcgcc
gtccttcgag 60 gaggtggcgc tcatgcccaa cggcagcttc aagaagatca gcctctctc ctacaagggc 120
aagtgggtcg tgctcttctt ctaccgctc gacttcacct tcgtgtgccc gacagaggtc 180 atcgcttct
ccgacagcgt gagtgcctt aacgagctca actgcgaggt cctcgcgtgc 240 tcgatagaca gcgagtacgc
gcacctgcag tggacgtgc aggaccgcaa gaaggcgcc 300 ctccggacca tggcgatccc aatgtagcc
gacaagacca agagcatcgc tcgttctac 360 ggcgtgctgg aggagagcca gggcgtggcc taccgcgct
tcttcatcat cgacccccat 420 ggcacgtcgc gtcagatcac cgtcaatgac atgccgtggt gccgcgagct
ggaggaggtt 480 ctacgcctgc tggaggctt tcagttcgtg gagaagcacg gcgaggtgtg cccgcgcaac
540 tggaagaagg gcgccccac gatgaagccg gaaccgaatg cgtctgtcga gggatacttc 600
agcaagcagg gatccatgga cgcaactgag ctgaagaaca aggggaacga agagttctcc 660 gccggccgct
atgtggaggc ggtgaactac ttctcaaagg cgatccagtt ggatgagcag 720 aacagtgtcc tctacagcaa
ccgctccgcc tgttttgag ccatgcagaa atacaaggac 780 gcgctggacg acgccgacaa gtgcatctcg
atcaagccga attgggcaa gggctacgtg 840 cgcgaggag cagctctcca tggcatgagc cgctacgacg
atgccattgc cgcgtatgaa 900 aaggggtca aggtggacc ttccaacagc ggctgcgcgc agggcgtaga
ggacgtgcag 960 gtacccaagg cccgcgaagc acgtgacccc atcgctcgcg tcttaccacc ggaggcgttc
1020 cgcaagatcc aagagaatcc caagctgtct ctacttatgc tgcagccgga ctacgtgaag 1080
atggtagaca ccgtcatccg cgacccttcg cagggccggc tgtacatgga agaccagcgc 1140 tttgccctga
cgctcatgta cctgagcggg atgaagattc ccaacgatgg tgatggcgag 1200 gaggaggaac gtccgtctgc
gaaggcgga gagacagcga agccaaaaga ggagaagcct 1260 ctaccgaca acgagaagga ggccctggcg
ctcaaggagg agggcaacaa gctgtacctc 1320 tcgaagaagt ttgaggaggc gctgaccaag taccaagagg
cgaggtgaa agacccaac 1380 aacactttat acattctgaa cgtgtcggcc gtgtacttcg agcagggtga
ctacgacaag 1440 tgcacgcgg agtgcgagca cggatcagc caggtcgcg agaaccactg cgactacaca
1500 atcattgca agctcatgac ccggaacgcc ttgtgcctcc agaggcagag gaagtacgag 1560
gctgctatcg acctttaca gcgcgccctt gtcgagtggc gtaaccctga caccctcaag 1620 aagctgacgg
agtgcgagaa ggagcaccaa aaggcggtg aggaagccta catcatcct 1680 gagatcgca agcagaaga
agacgaaggt aaccagtact tcaaggagga taagttcccc 1740 gaggccgtgg cagcgtacac ggaggccatc
aagcgcaacc ctgccgagca cacctcctac 1800 agcaatcgcg cggcccgcta catcaagctt ggagccttca
acgacgccct caaggacgcg 1860 gagaagtgca ttgagctgaa gcccgacttt gttaagggct acgcgcgcaa
gggtcatgct 1920 tacttttga ccaagcagta caaccgcgcg ctgcaggcgt acaatgaggg cctcaagggtg
1980 gaccgagca atgcggactg caaggatggg cggatcgcga caatcatgaa gattcaggag 2040
atggcatctg gccaatccgc ggatggcgac gaggcgccgc gccgggccat ggacgatcct 2100 gaaatcgcg
caatcatgca agatagctac atgcaactag tgttgaagga gatgcagaac 2160 gatccacgc gcattcagga
gtacatgaag gactccggga tctcatcgaa gatcaacaag 2220 ctgatttcag ctggcatcat tcgttttgg
caggaattca gcctgacgga cccggcggtg 2280 ctgggcgagg agactcacct gcgcgtccgc gtggtgccg
acaaggcgaa caagacgctg 2340 acggtggagg ataacggcat cggcatgacc aaggcggacc tcgtgaacaa
tctgggcacg 2400 atcgcgct cccgcacgaa ggcgttcacg gaggcactgg aggcggcgcg cgacatgagc
2460 atgatcgcc agttcggtg cggttctac tccgcgtacc ttgtggcgga ccgcgtgacg 2520
gtggtgtcga agaacaactc ggacgaggcg tacgtatggg agtcgtccgc gggcggcacg 2580 ttcaccatca
cgagcgtgcc ggagtcggac atgaagcgcg gcacgcgat cacgtgcac 2640 ctaaaggagg accagcagga
gtacctggag gagcgccggg tgaaggagct gatcaagaag 2700 cactccgagt tcatcgcta cgacatcgag
ctgatgggtg agaagcggc ggagaaggag 2760 gtgacggagc aggcagga ggaggcag tcaagaaga
agtcctgcgg ggacgaggcg 2820 gagccgaagg tggaggaggt gacggaggc ggcgaggaca agaagaaga
gacgaagaag 2880 gtgaaggagg tgacgaagac gtacgaggtc cagaacaagc acaagcgct ctggacgcg
2940 gaccgaagg acgtgacgaa ggaggagtac gcggccttct acaaggccat ctccaacgac 3000
tgggaggacc cggcgcgac gaagcacttc tcggtggagg gccagctgga gttccgcgcg 3060 atcgcgcttcg
tgccgaagcg cgcgcgttc gacatgttcg agccgaacaa gaagcgcaac 3120 aacatcaagc tgtacgtgcg

ccgcgtgttc atcatggaca actgcgagga cctgtgcccc 3180 gactggctcg gcttcgtgaa gggcgctcgtg
gacagcgagg acctgccgct gaacatctcg 3240 cgcgagaacc tgcagcagaa caagatcctg aaggtgatcc
gcaagaacat cgtgaagaag 3300 tgcctggagc tgctcgaaga gatagcggag aacaaggagg actacaagca
gttctacgag 3360 cagttcggca agaacatcaa gctgggcatc cacgaggaca cggcgaaccg caagaagctg
3420 atggagttgc tgcgttcta cagcaccgag tcgggggagg agatgacgac actgaaggac 3480
tacgtgacgc gcatgaagcc ggagcagaag tcgatctact acatcactgg cgacagcaa 3540 aagaagctgg
agtcgtcgcc gttcatcgag aaggcgagac gctgcgggct cgaggtgctg 3600 ttcatgacgg agccgatcga
cgagtacgtg atgcagcagg tgaaggactt cgaggacaag 3660 aagttcgcgt gcctgacgaa ggaaggcgtg
cacttcgagg agtccgagga ggagaagaag 3720 cagcgcgagg agaagaaggc ggctgcgag aagctgtgca
agacgatgaa ggaggtgctg 3780 ggcgacaagg tggagaaggt gaccgtgtcg gagcgctgt cgacgtcgcc
gtgcatcctg 3840 gtgacgtcgg agtttggtg gtcggcgac atggaacaga tcatgcgaa ccaggcgctg
3900 cgcgactcca gcatggcgca gtacatggtg tccaagaaga cgatggaggt gaaccccgac 3960
caccatca tcaaggagct gcgcgcgcg gtggaggcgg acgagaacga caaggccgtg 4020 aaggacctcg
tcttctgct cttcgacacg tcgctgctca cgtccggctt ccagctggat 4080 gacccaccg gctacgccga
gcgcacaa cgcatgatca agctcggcct gtcgctcgac 4140 gaggaggagg aggaggtcgc cgaggcgccg
ccggccgagg cagccccgc ggagtcacc 4200 gccggcacct ccagcatgga gcaggtggac gatatcatgg
cgcagaatga taagatcgcc 4260 cccaggacc aggactcctt cctcgatgac cagcccgcg ttcgccgat
cccgtccttc 4320 gacgacatgc cgctgcacca gaacctgctg cgtggcatct actcgtagc gttcgagaag
4380 ccgtccagca tccagcagcg cgcatagcc cccttcacgc gcggcgcgca catcatcgcg 4440
caggccagt ccgtaccgg caagacgggt gccttctcca tcggtctgct gcagcgctg 4500 gacttccgcc
acaacctgat ccagggcctc gtgctctccc cactcgca gctggccctg 4560 cagacggcg aggtgatcag
ccgcatcggg gagttcctgt cgaacagctc caagtctgc 4620 gagaccttg tcggcgccac gcgctgca
gatgacctg gcaagtgcga gccggcgctc 4680 atcgttgcg tgggcacgcc gggccgctg tccgacgtga
tcaagctgg cgcgctgcg 4740 acagagtgc tcgctgtgt ggtgctcgac gaggtgatg agatgctgtc
tcagggttc 4800 gcggaccaga ttacgagat ctccgcttc ctgccgaagg acatccagg cgctcttc
4860 tccgccacga tcccgaggga ggtactggag ctgacgaaga agttcatgcg cgactaa 4917 <200>
SEQUENCE CHARACTERISTICS: <210> SEQ ID NO 101 <211> LENGTH: 2735 <212> TYPE: DNA
<213> ORGANISM: Artificial Sequence <220> FEATURE: <223> OTHER INFORMATION: DNA
sequence encoding fusion (poly-protein) constructs comprising multiple Leishmania
antigens <400> SEQUENCE: 101 catatgcacc accaccacca ccacatgtcc tgcgtaacg ccaagatcaa
ctctcccgcg 60 ccgtccttcg aggaggtggc gctcatgccc aacggcagct tcaagaagat cagcctctcc 120
tcctacaagg gcaagtgggt cgtgctcttc ttctacccgc tcgacttcac ctctgtgtg 180 ccgacagagg
tcatcgctt ctccgacagc gtgagtcgct tcaacgagct caactgcgag 240 gtcctcgct gctcgataga
cagcgagtac gcgcacctgc agtgagcgt gcaggaccgc 300 aagaagggcg gcctcgggac catggcgatc
ccaatgctag ccgacaagac caagagcatc 360 gtcgttcct acggcgtgct ggaggagagc cagggcgtgg
cctaccgcgg tctcttcac 420 atcgaccccc atggcatgct gcgtcagatc accgtcaatg acatgccggg
ggccgcagc 480 gtggaggagg ttctacgcct gctggaggct tticagttcg tggagaagca cggcgagggtg
540 tgcccccgca actggaagaa gggcgcccc acgatgaagc cggaaccgaa tgcgtctgtc 600
gagggatact tcagcaagca gggatccatg gacgcaactg agctgaagaa caaggggaac 660 gaagagttct
ccgcggccg ctatgtggag gcgtgaact acttctcaaa ggcgatccag 720 ttggatgagc agaacagtgt
cctctacagc aaccgctccg cctgttttgc agccatgcag 780 aaatacaagg acgcgtgga cgacccgac
aagtgcatt cgatcaagcc gaattgggccc 840 aagggtacg tgcggcgagg agcagctctc catggcatgc
gccgtacga cgtgccatt 900 gccgcgtatg aaaaggggct caaggtggac ccttccaaca gcggctgcg
gcagggcgtg 960 aaggacgtgc aggtagccaa gggccgcgaa gcacgtgacc ccacgtcgc cgtcttcacc
1020 ccggaggcgt tccgcaagat ccaagagaat cccaagctgt ctctacttat gctgcagccg 1080
gactacgtga agatggtaga caccgtcatc cgcgaccctt cgaggggcg gctgtacatg 1140 gaagaccagc
gctttgccct gacgtcatg tacctgagcg gaatgaagat tcccaacgat 1200 ggtgatggcg aggaggagga
acgtccgtct gcgaaggcg cagagacagc gaagccaaaa 1260 gaggagaagc ctctcaccga caacgagaag
gaggccctgg cgctcaagga ggagggaac 1320 aagctgtacc tctcgaagaa gtttgaggag gcgctgacca
agtaccaaga ggcgcaggtg 1380 aaagaccca acaacacttt atacattctg aacgtgtcgg ccgtgtactt
cgagcagggt 1440 gactacgaca agtgcacgc cgagtgcgag caggtatcg agcacggctg cgagaaccac
1500 tgcgactaca caatcattgc gaagctcatg acccggaacg cttgtgcct ccagggcag 1560
aggaagtacg aggtgctat cgacctttac aagcgcgccc ttgtcgagtg gcgtaacct 1620 gacacctca
agaagctgac ggagtgcgag aaggagcacc aaaaggcggt ggagggaagc 1680 tacatcgatc ctgagatcgc
gaagcagaag aaagacgaag gtaaccagta cttaaggag 1740 gataagttcc ccgaggccgt ggcagcgtac
acggaggcca tcaagcgca ccctgccgag 1800 cacacctcct acagcaatcg cgcgcccgcg tacatcaagc
ttggagcctt caacgacgcc 1860 ctcaaggacg cggagaagtg cattgagctg aagcccgact ttgttaaggg
ctacgcgcg 1920 aagggtcatg ctacttttg gaccaagcag tacaaccgcg cgctgcaggc gtacaatgag
1980 ggcctcaagg tggaccgcg caatgcggag tgcaaggatg ggcggtatcg cacaatcatg 2040
aagattcagg agatggcatc tggccaatcc gcggatggcg acgaggggcg gcgcggggcc 2100 atggacgatc
ctgaaatcgc ggcaatcatg caagatagct acatgcaact agtgtgaag 2160 gagatgcaga acgatccac
gcgcattcag gattacatga aggactccg gatctcatcg 2220 aagatcaaca agctgatttc agtggcatc
attcgtttt gtcaggaatt ctgcagatat 2280 ccacacact ggcggcgct cgagcagatc cggctgctaa
caaagcccga aaggaagctg 2340 agttggctgc tgccaccgct gagcaataac tagcataacc ccttggggcc

tctaaacggg 2400 tcttgagggg ttttttgctg aaaggaggaa ctatatccgg ataattcttg aagacgaaag
2460 ggctcgtga tacgcctatt tttataggtt aatgtcatga taataatggt ttcttagacg 2520
tcaggtggca cttttcgggg aaatgtgcgc ggaacccta tttggttatt tttctaaata 2580 cattcaaata
tgtatccgct catgagacaa taaccctyga taaatgcttc aataatattg 2640 aaaaaaggaa gaatatgaag
tatttcaaca tttcccggt ccccttatt ccttttttt 2700 gccgccattt tgcctttctg tttttggttc
accca 2735

Other Reference Publication (3):

Campos-Neto et al., "Cloning and expression of Leishmania donovani gene instructed by a peptide isolated from major histocompatibility complex class II molecules of infected macrophages," Journal of Experimental Medicine 182(5):1423-1433, 1995.

Other Reference Publication (5):

Coulson et al., "Differential expression of Leishmania major beta-tubulin genes during the acquisition of promastigote infectivity," Mol Biochem Parasitol 82 (2):227-236, Nov. 1996.

Other Reference Publication (6):

De Andrade et al., "Recombinant Leishmania Hs90 and Hsp70 are recognized by sera from visceral leishmaniasis patients but not Chagas' disease patients," Journal of Clinical Microbiology 30(2):330-335, 1992.

Other Reference Publication (7):

Dillon et al., "Characterization of a Leishmania tropica antigen that detects immune responses in desert storm viscerotropic leishmaniasis patients," Proc. Natl. Acad. Sci USA 92:7981-7985, 1995.

Other Reference Publication (8):

Fong and Lee, "Beta tubulin gene of the parasitic protozoan Leishmania mexicana," Mol Biochem Parasitol 31(1):97-106, Oct. 1988.

Other Reference Publication (10):

GenBank Accession No. AC003679, "Leishmania major chromosome 1, complete sequence," Mar. 24, 1999.

Other Reference Publication (11):

GenBank Accession No. U73845, "Leishmania major protein antigen LmSTI1 mRNA," Dec. 3, 1996.

Other Reference Publication (14):

Mougneau et al., "Expression cloning of a protective Leishmania antigen," Science 268:563-566, 1995.

Other Reference Publication (16):

Osland et al., "Isolation and characterization of recombinant antigens from Leishmania aethiopica that react with human antibodies," Infection and Immunity 60 (4):1368-1374, Apr. 1992.

Other Reference Publication (17):

Pir2 Database Accession No. S54162, "Leishmania donovani," Jul. 8, 1995.

Other Reference Publication (18):

Shapira and Pedraza, "Sequence analysis and transcriptional activation of heat shock protein 83 of Leishmania mexicana amazonensis," Molecular and Biochemical Pathology 42(2):247-255, 1990.

Other Reference Publication (19):

Skeiky et al., "A recombinant Leishmania antigen that stimulates human peripheral blood mononuclear cells to express a Th1-type cytokine profile and to produce interleukin 12," Journal of Experimental Medicine 181(4):1527-1537, 1995.

Other Reference Publication (20):

Sheiky et al., "Proliferative and cytokine responses of human PBMC to cloned Leishmania braziliensis heat shock and ribosomal antigens," Journal of Immunology

158(8 pt. 2):93A, Abstract #517, 1993.

Other Reference Publication (21):

Webb et al., "Human and murine immune responses to a novel Leishmania major recombinant protein encoded by members of a multicopy gene family," Infection and Immunity 66(7):3279-3289, Jul. 1998.

Other Reference Publication (22):

Yang et al., "Identification and characterization of host-protective T cell epitopes of a major surface glycoprotein (gp63) from Leishmania major," Immunology 72(1):3-9, Jan. 1991.

Other Reference Publication (23):

Afonso et al., The adjuvant effect of interleukin-12 in a vaccine against Leishmania major, Science 263:235-237, Jan. 14, 1994.

Other Reference Publication (25):

Burns et al., "Molecular characterization of a kinesin-related antigen of Leishmania chagasi that detects specific antibody in African and American visceral leishmaniasis," Proc. Natl. Acad. Sci. USA 90:775-779, Jan. 1993.

Other Reference Publication (26):

Champsì and McMahon-Pratt, "Membrane glycoprotein M-2 protects against Leishmania amazonensis infection," Infection and Immunity 52(12):3272-3279, Dec. 1988.

Other Reference Publication (27):

Chatelain et al., "IL-4 induces a Th2 response in Leishmania major-infected mice," The Journal of Immunology 148(4):1182-1187, Feb. 15, 1992.

Other Reference Publication (29):

Flinn et al., "Expression of a hydrophilic surface protein in infective stages of Leishmania major," Molecular and Biochemical Parasitology 65:259-270, 1994.

Other Reference Publication (31):

Pan et al., "Developmental life cycle of Leishmania--cultivation and characterization of cultured extracellular amastigotes," The Journal of Eukaryotic Microbiology 40(2):213-223, Mar.-Apr. 1993.

Other Reference Publication (32):

Reiner and Locksley, "The regulation of immunity to Leishmania major," Annu. Rev. Immunol. 13:151-177, 1995.

Other Reference Publication (33):

Rodrigues et al., "Selective inability of spleen antigen presenting cells from Leishmania donovani infected hamsters to mediate specific T cell proliferation to parasite antigens," Parasite Immunology 14(1):49-58, Jan. 1992.

Other Reference Publication (34):

Skeiky et al., "Antigens shared by Leishmania species and Trypanosoma cruzi: immunological comparison of the acidic ribosomal P0 proteins," Infection and Immunity 62(5):1643-1651, May 1994.

[Previous Doc](#)

[Next Doc](#)

[Go to Doc#](#)